THE INTERACTIVE EFFECTS OF GENETICS AND CARDIORESPIRATORY FITNESS LEVEL ON COGNITIVE PERFORMANCE IN HEALTHY OLDER ADULTS

BY

JENNIFER KIM

DISSERTATION

Submitted in partial fulfillment of the requirements
for the degree of Doctor of Philosophy in Neuroscience
in the Graduate College of the
University of Illinois at Urbana-Champaign, 2016

Urbana, Illinois

Doctoral Committee:

Professor Arthur Kramer, Chair
Professor Neal Cohen
Associate Professor Bettina Francis
Professor Edward McAuley

ABSTRACT

This study was conducted as an exploratory analysis of the potential interaction between genetics and cardiorespiratory fitness level on cognition in healthy older adults. The main questions were 1) can genetic status moderate the effect of cardiorespiratory fitness level on cognition and 2) can cardiorespiratory fitness level influence the effect of genetic status on cognition.

A cross-sectional sample of healthy older adults were recruited for a longitudinal randomized controlled exercise intervention. Participants were admitted into the study based on their sedentary lifestyle and this analysis was conducted on data collected at the outset of the intervention.

Dopamine-related genes were chosen based on dopamine's influence on cognition and its sensititvy to aging. The Catechol-O-Methyltransferase (COMT) and Dopmaine Beta Hydroxylase (DBH) genes were chosen because their protein products influence levels of dopamine within the brain. Further, each gene has two common functional single nucleotide polymorphisms which effect the functionality of the protein product.

Two growth factor genes, Insulin-like Growth Factor I and Brain Derived Neurotrophic Factor, were chosen based on the role they play in brain development and overall health as well as their moderation by exercise.

The primary measure of cardiorespiratory fitness was maximal oxygen uptake (VO_2) and neuropsychological assessments were collected either through computer-based or paper-and-pencil methods.

Overall, results suggest that genetic status did interact with cardiorespiratory fitness level to influence both working and spatial memory as well as various aspects of executive functioning including response inhibition, maintenance and coordination of multiple sets, and cognitive flexibility.

Results of the study point to the need for approaches beyond single gene assocations such as genome wide association studies (GWAS), epigenetic analyses, gene-gene interactions and interactions with environmental factors to take into consideration all the factors that contribute to the variability in the cognitive aging process.

Dedicated to all the people who never stopped believing in me.

"Life is not easy for any of us. But what of that? We must have perseverance and above all confidence in ourselves. We must believe that we are gifted for something and that this thing must be attained."

-Marie Curie

Table of Contents

Chapter 1: Introduction

It is well known that many changes occur in human cognition over the lifespan. Unfortunately, after the age of 30, many of these changes are declines as opposed to improvements. For example, research has shown aging-related declines in several aspects of cognition along with concominant declines in brain structure, function, and neurochemistry (Hedden & Gabrieli, 2004; Strong, 1998; Yankner, Lu, & Loerch, 2008; West, 1996). While there exists a global aging-related decline, there are still individual variations in the degree to which these declines occur since many factors contribute to these changes.

In this thesis, I will focus on two specific factors that have both been implicated in contributing to individual differences in many aspects of aging-related declines – cardiorespiratory fitness and genetics. As a lifestyle factor, increase of cardiorespiratory fitness level has been associated with healthier aging and reduced age-related declines in brain structure, function, and cognition (Colcombe, Kramer, McAuley, Erickson, & Scalf, 2004; Erickson & Kramer, 2009; Gomez-Pinilla & Hillman, 2013; Lista & Sorrentino, 2010; Mcauley, Kramer, & Colcombe, 2004; Prakash, Voss, Erickson, & Kramer, 2015). Several studies have shown that aerobic exercise interventions can ameliorate or even improve different types of age-related declines by increasing participants' cardiorespiratory fitness levels (Colcombe & Kramer, 2003; Colcombe et al., 2004a, 2006; Etnier et al., 1997). Cross-sectional research has also shown that older adults who are aerobically fit generally perform better on cognitive tasks compared to those less fit (Voss et al., 2010; Erickson et al., 2009b; Colcombe et al., 2003, 2004a).

Genetics is a major biological determinant of cognitive abilities. Although everyone has the same general genetic makeup, there are small nuances in the genetic code that can cause differences in the availability and functionality of proteins in the body. Single nucleotide polymorphisms (SNPs) occur in the human genome and contribute to almost 90% of human genetic variation. These SNPs can cause individuals to have different versions of the same genes, and depending on location, can translate to more or less of a particular protein. During the early and middle years of human development, these variations may or may not have detectable phenotypes; however as aging takes its toll,

differences can become more apparent. For instance, Nagel et al. (2008) found that a SNP on a dopamine gene (specifically the Catechol-O-Methyltransferase gene) had a genetic effect on cognition in older adults, but not middle-aged adults, providing some evidence that genetic effects may be magnified with aging.

The majority of studies have investigated the influence of cardiorespiratory fitness and genetics on cognition in healthy older adults independently, but a growing number have begun looking at an interactive effect (e.g. Deeny et al., 2008; Erickson et al., 2013; Etnier et al., 2007, 2015; Schuit, Feskens, Launer, & Kromhout, 2001; Stroth et al., 2010; Voelcker-Rehage, Jeltsch, Godde, Becker, & Staudinger, 2015). The possibility exists that the influence of cardiorespiratory fitness levels may outweigh genetic effects, or that maintenance of cardiorespiratory fitness levels may be more important for some individuals than others. In this thesis, I explore the interactive effect of cardiorespiratory fitness level and genetics on cognitive abilities in a cross-sectional sample of healthy older adults.

In the following sections I first review the literature on age-related structural, functional, and neurochemical brain changes, followed by how these changes relate to behavior since behavior is the outcome of interest. The effects of cardiorespiratory fitness and genetics are briefly reviewed here and will be discussed in further detail in individual chapters.

Age-related Declines

Structural and functional brain changes. There is evidence for a general age-related reduction in gross brain volume with substantially greater declines in the prefrontal and frontal regions in comparison to sensory cortical regions (Cabeza, 2001a; Colcombe, Kramer, Erickson, & Scalf, 2005; Raz et al., 1997, 2005). This reduction in brain volume, at least for older adults without a diagnosis for age-associated neuropathologies such as Alzheimer's disease, does not appear to be an actual loss of neurons, but reductions in synaptic integrity and density (Hof & Morrison, 2004; Morrison & Hof, 2007), volumetric and structural white matter changes (Colcombe et al., 2004a; O'Sullivan et al., 2001), loss of dendritic processes, and reductions in the number of synapses (West, 1996). Postmortem studies have shown that there is a slow but persistent rate of decline in overall brain

weight and volume of about 2% per decade (Cabeza, 2001b). However, using an *in vivo* MRI method, Raz et al. (1997) found an average rate of decline of 4.9% per decade for prefrontal gray matter and the correlation between age and prefrontal volume was significantly larger than with any other brain area. Although polymodal and visual association cortices do exhibit sizeable age-related declines, the prefrontal cortex (PFC) shows the most sensitivity to age, while primary sensory (Raz et al., 2005) and motor areas remain relatively stable throughout the lifespan (Raz et al., 1997). Further, while marked volume loss in the PFC occurs with normal healthy aging, similar losses in the medial temporal lobes and hippocampus are often indicators of pathologic aging (Cabeza, 2001b; Raz, 2000; Yankner et al., 2008). Medial temporal and hippocampal volume losses have also been observed in healthy aging populations, but to a much lesser extent than pathologic populations (Jack et al., 1998). Although evidence supports a general aging-related structural decline in the brain, there exists a large amount of individual variability (Raz et al., 2005).

Advent of neuroimaging techniques such as functional magnetic resonance imaging (fMRI) and positron emission tomography (PET) has marked the cornerstone of our understanding of the functional changes in neural recruitment that might be associated with healthy aging. One of the common observations in several functional imaging studies is the additional recruitment of neuronal resources in the prefrontal cortices of older adults. For example, Reuter-Lorenz et al. (2000) found that during both a verbal and spatial working memory task on which older and young adults had high accuracy, older adults showed bilateral anterior activations, while young adults showed unilateral activations (i.e. left for verbal and right for spatial). The authors concluded that that the reduced anterior lateralization in older adults was compensatory, such that additional recruitment reflected an augmentation of task performance. Cabeza (2002) formalized this observed pattern into a model called hemispheric asymmetry reduction in older adults (HAROLD). However, there is still debate as to whether the HAROLD pattern of activation is compensatory or an indication of decline. For example, Colcombe et al. (2005) reported significantly greater bilateral PFC activation for poor-performing compared to good-performing older adults during inhibition on incongruent trials of a Flanker task. The authors suggest that bilateral activations are only compensatory to the degree that the functions subserved by the

recruited areas play a complementary role in task performance. This finding also points to the existence of individual variability in the functional changes associated with healthy aging.

In general, the extant literature points to the main fact that changes in brain structure and function are pervasive during aging, particularly in the PFC. And although general declines are observed, there is a great deal of individual variability.

Cognitive changes. The amount of cognitive decline associated with aging varies with the type of cognitive domain being tested. For example, crystallized cognitive abilities such as verbal and numeric abilities, autobiographical memory, and emotional processing remain relatively stable or even improve throughout the lifespan. However, fluid abilities such as processing speed, working memory, and executive control decline with age and the decline accelerates close to death (Hedden & Gabrieli, 2004; Park & Gutchess, 2002). The age-sensitivity of executive control is of particular interest because of its importance to real world functioning. Executive control refers to cognitive processes associated with planning, coordination, and monitoring of other cognitive operations (i.e. a central executive; Baddeley, 1992, 2010; Salthouse & Miles, 2002). This decline in executive control is associated with the concomitant aging-related structural and functional changes of the prefrontal cortex (PFC; discussed above), which is known to subserve, in part, executive control processing (Raz et al., 2005; West, 1996).

There are many aspects of executive control and many different tasks that can be used to assess the integrity of these different aspects of executive control processing. For example, one facet of executive control is active mental manipulation as opposed to passive storage of relevant information, often referred to as updating (Baddeley, 1992, 1996, 2010; Johnson, Raye, Mitchell, Greene, & Anderson, 2003; Langley & Madden, 2000; Miyake et al., 2000). Short-term passive storage is often preserved in older adults (Hedden & Gabrieli, 2004; Park & Gutchess, 2002), whereas the ability to manipulate the stored information declines with age (Chen & Li, 2007; Van der Linden, Brédart, & Beerten, 1994). Updating can be tested by asking participants to monitor and code incoming information for task relevance, and then continuously manipulate, evaluate, and replace old, irrelevant content with newer, more relevant information to successfully complete the task (Chen & Li, 2007). In 2007, Chen & Li used separate measures of updating, processing speed, and fluid

intelligence to show that increased age is associated with reductions in updating ability, and that this reduced efficiency is associated with poorer performance on fluid intelligence tests. They found that older participants were slower on speed tasks, performed poorer on fluid intelligence assessments, and had a decreased percentage of correct responses in updating tasks (e.g. running memory task). More importantly, using a series of hierarchical regression analyses, they verified that age-related differences in updating variables was at least partially independent of the age-related differences in speed variables. Furthermore, they created a structural equation model with good convergent validity of each of their constructs (i.e. speed, updating, and fluid intelligence) and good discriminate validity between the speed and updating constructs after controlling for age. This model showed large path coefficients from age to both speed and updating, indicating an association between increased age and slower processing speed and poorer updating efficiency. In addition, the model provided evidence that updating mediates (at least in part) age-related effects on fluid intelligence.

Another aspect of executive control is the ability to maintain and coordinate multiple task goals, which I will refer to as coordination. Two main aspects of coordination are time-sharing and shifting. Time-sharing refers to the effectiveness of coordinating two simultaneous tasks, distinct from the efficiency of performing each task separately (Salthouse & Miles, 2002). Shifting refers to the ability to maintain and coordinate a set of task goals in order to shift between one and another. In simplest terms, shifting involves disengaging an irrelevant task goal, followed by active engagement of a relevant task goal (Kramer, Hahn, & Gopher, 1999; Miyake et al., 2000). In the laboratory, time-sharing and shifting are often assessed using dual task and task-switching paradigms respectively. Briefly, dual task paradigms require participants to perform two tasks simultaneously (e.g. vowel/consonant letter judgment and odd/even number judgment) while task-switching paradigms require participants to switch between two different tasks (e.g. vowel/consonant letter judgment followed by an odd/even number judgment). These types of tasks are also referred to as tasks of divided attention.

In 2002, Salthouse & Miles had young and old adults perform three different primary tasks separately and in concurrence with a common secondary task. They found significant correlations among time-sharing costs in all three dual task conditions,

providing evidence for a separable time-sharing construct. Furthermore, the time-sharing factor was distinct from factors representing perceptual speed and other cognitive factors, providing more evidence of a distinct executive factor. This time-sharing factor was correlated with age such that increasing age led to increasing disruption in time-sharing. Increased age was also associated with lower levels of performance in each task separately and concurrently, even after single task accuracy was adjusted for across age. Additionally, two separate meta-analyses of tasks of divided attention found significant age differences above and beyond the effects of general slowing (Verhaeghen & Cerella, 2002; Verhaeghen, Steitz, Sliwinski, & Cerella, 2003).

In 1999, Kramer, Hahn, and Gopher utilized three different versions of a task-switching paradigm and found that switch trial reaction times were significantly longer for older adults than young adults. These effects were robust across a range of preparation intervals, different levels of working memory load, and whether or not the switch was predictable. They also found that older adults made more perseverative errors on the Wisconsin Card Sorting Test (WCST), indicating impairment in the ability to shift from one rule to another and in the ability to stop the use of one rule and initiate the use of another.

This leads into the final aspect of executive control discussed here – the ability to inhibit dominant or automatic responses when necessary or to actively suppress one response in order to make another (Miyake et al., 2000). Older adults show deficits in tasks requiring this type of inhibitory processing. One of the most commonly used tasks to assess inhibitory executive control is the Stroop task. In the Stroop task, participants are presented with color words printed in different ink colors. They are then asked to report on the ink color and not the meaning of the word. In general, performance of this task is poorer when the color word and the ink color are in conflict – the Stroop effect. This task utilizes the difficulty of inhibiting the automaticity of reading (i.e. task irrelevant information) in order to perform the weaker task-relevant reporting of the ink color (Stroop, 1935). In 1999, Brink and McDowd conducted an experiment comparing the age effects of quantitative (i.e. variations in number of choices of colors) versus qualitative (i.e. name the ink color of a block versus name the ink color of a color word) measures. By analyzing proportional Stroop effect scores to control for general age-related slowing, they found that age did not interact with choice, but did interact with task type such that the

young and old performed comparably on the Color-Block Task, but the young outperformed the old in the Color-Word Task. This shows that the effect of generalized slowing cannot fully explain the age differences in the performance on the two tasks and that inhibitory processing is disproportionately affected by age.

The Flanker task is also commonly used to test inhibitory executive processing. In the Flanker task, participants are shown five arrows and asked to report on the direction of the central arrow. There are two conditions to this task. In the congruent condition, the flanking arrows are in the same direction (<<<<<) as the central arrow. In the incongruent condition, flanking arrows are in the opposite direction (<<><<), and participants must inhibit the irrelevant distracting information of the flanking arrows in order to make a correct response. Colcombe et al. (2005) reported that older adults were reliably slower to respond to both congruent and incongruent trials compared to young adults. More importantly, the older adults were proportionally more disrupted by incongruent trials than young adults when proportional interference scores were assessed (i.e. [incongruent response time – congruent response time]/congruent response time). Also, there was no reliable correlation between congruent response time and proportional interference scores, showing that the main deficit was in inhibitory processing of incongruent items and not a result of a main effect of slowed response time.

Taken together, evidence points to a general age-related deficit in executive control processing. Declines in the ability to update memory stores, coordinate multiple tasks, and inhibit irrelevant information can have severe repercussions on a person's quality of life during aging. Therefore, it is important to understand and explore the different factors that contribute to or alleviate this age-related cognitive decline.

Cardiorespiratory Fitness

Lifestyle factors are increasingly being recognized as simple ways of promoting physical and cognitive health. There is an ever growing literature examining the physical, cognitive, and neural changes associated with physical activity, cardiorespiratory fitness, and exercise, and how these may contribute to brain health. The non-human animal literature points to cardiorespiratory fitness as a factor that can contribute to individual differences in age-related cognitive, neurochemical, and structural differences. The

beneficial effects of exercise may act directly on the molecular machinery of the brain itself by influencing neurotrophic factors (Cotman & Berchtold, 2002) and neurotransmitters such as dopamine. For example, (Gilliam et al., 1984) found increased D2 receptor binding within the striatum in young exercised rats. Further, MacRae, Spirduso, Walters, Farrar, & Wilcox (1987) found exercise diminished age-related declines in dopamine metabolism and D2 receptor density.

Additionally, Neeper, Gómez-Pinilla, Choi, & Cotman (1995) found that voluntary wheel running increased levels of BDNF mRNA in the rat hippocampus. These increased mRNA levels were observed within days in both male (Neeper, Gómez-Pinilla, Choi, & Cotman, 1996) and female (Berchtold, Kesslak, Pike, Adlard, & Cotman, 2001) rats and levels were sustained after several weeks of exercise. Further, there was also a parallel increase in BDNF protein. Additional studies have found that voluntary wheel running, above and beyond just an enriched environment, leads to neurogenesis, increased cell survival, and improved spatial navigation (Kobilo et al., 2011; van Praag, Christie, Sejnowski, & Gage, 1999; van Praag, Kempermann, & Gage, 1999), and physical activity can promote brain vascularization (Black, Isaacs, Anderson, Alcantara, & Greenough, 1990).

In older humans, baseline measures of cardiorespiratory fitness (i.e. VO_2 peak) have been positively associated with preserved cognitive function over a period of six years (mean age at baseline = 69; Barnes, Yaffe, Satariano, & Tager, 2003). In addition, several cross sectional studies have found that higher levels of cardiorespiratory fitness can attenuate age-related cognitive declines (e.g. Hillman, Weiss, Hagberg, & Hatfield, 2002; Renaud, Bherer, & Maquestiaux, 2010; Spirduso, 1975). Supporting these cognitive findings, increased levels of cardiorespiratory fitness have also been associated with increased brain volume specifically in the frontal and temporal cortices (Colcombe et al., 2003, 2006), as well as hippocampi in older adults (Erickson et al., 2009b).

Further evidence comes from randomized controlled trials in which relatively sedentary older adults, usually over the age of 60, are randomized into an aerobic training group (e.g. walking) or a non-aerobic control group (e.g. stretching and toning). These studies have presented mixed results due to differences in cardiorespiratory fitness assessment, but in general show that exercise induced increases in cardiorespiratory fitness positively influence cognition (Kramer & Erickson, 2007). Further, in a meta-

analysis of aerobic fitness training studies with older adults, Colcombe & Kramer (2003) found that aerobic exercise training increased performance, regardless of the type of cognitive task, training method, or participant characteristics. In a more recent meta-analysis, Smith et al. (2010) corroborated the Colcombe & Kramer meta-analysis, specifically finding modest effects of aerobic fitness training on attention and processing speed, executive function, and memory. Although aerobic exercise training has relatively broad effects on perceptual and cognitive processes, the benefits seem to be largest for executive control processes (Colcombe & Kramer, 2003; Kramer & Erickson, 2007). The literature also suggests that 6 months of moderate aerobic exercise is sufficient to show benefits on cognitive function, with the greatest benefits again on executive control processes (Erickson & Kramer, 2009). Specifically, Colcombe, Kramer, Erickson, et al. (2004) found that healthy older adults that were aerobically trained for 6 months showed improved inhibitory processing in a Flanker task and a change in the pattern of fMRI activation, becoming more similar to that displayed by younger controls. In another study, Colcombe et al. (2006) showed that walking three days a week for ~1 hour per day for six months increased frontal and temporal gray matter volume, as well as anterior white matter volume, in healthy older adults. Overall, the take home message is that physical activity, exercise and cardiorespiratory fitness show a positive relationship with cognition in healthy older adults.

Genetics

Genetic factors also contribute to variability in the myriad of aging-related declines. Twin studies have contributed to the understanding of the extent to which genetics versus environmental factors play a role in various aspects of human behavior, and how these contributions may change with aging. For example, a study of male twins between 69 and 80 years old reported a heritability of 79% for a latent trait of executive control (Swan & Carmelli, 2002). Further, in a review of several twin studies from various centers, Lee, Henry, Trollor, & Sachdev (2010) found genetic influence on processing speed and executive functions seems to increase with increasing age; and further, that environmental factors seem to show increased influence after the age of 65. This implicates the possibility

of using external resources, such as exercise interventions, to potentially rescue genetically predetermined behavioral outcomes in older age.

A person's genetic makeup can influence how much a certain neurochemical is made along with its availability and distribution, and in turn affect brain structure and function. Although general declines in brain structure and function are observed with aging, one's genetic makeup can influence the effect these declines will have on cognition. Allelic association has been utilized to investigate the role of particular genes in specific behaviors by inferring neurochemical brain changes.

Allelic association is an approach that integrates the fields of cognitive neuroscience and molecular genetics. This technique finds its roots in studies examining the heritability of certain aspects of cognition, mostly twin studies (Bouchard, Lykken, McGue, Segal, & Tellegen, 1990; Fan, Wu, Fossella, & Posner, 2001). Since the human genome cannot be experimentally manipulated, the target for such studies is naturally occurring genetic variation responsible for some phenotype – the more specific the better. Because allelic association studies look for correlations between alleles in a population as opposed to within families, the power to detect genes with a small effect size is greatly increased (Liu et al., 1996; Plomin & Craig, 1997).

Harris & Deary (2011) define a candidate gene study as "a study that investigates associations between genetic variants in a gene suspected of influencing a trait of interest (by virtue of the function or chromosomal location of the gene) and the trait." At the outset of this approach it is important to choose candidate genes with a well-defined measurable phenotype. Most studies focus on candidate genes with functional markers that result in amino acid coding differences or have a physiological effect (Deary, Wright, Harris, Whalley, & Starr, 2004; Goldberg & Weinberger, 2004; Greenwood & Parasuraman, 2003; Mattay & Goldberg, 2004; Parasuraman, Greenwood, Kumar, & Fossella, 2005; Plomin & Craig, 1997). These genes are chosen by how the gene products play a molecular and cellular role in areas of the brain involved in specific types of behavior or disease. Allelic variation arises from small differences that occur in the chain of nucleic acids making up the gene. The most common variation is called a single nucleotide polymorphism (SNP), defined as "a position on a chromosome where humans are known to differ with regard to which base pair occurs" (Harris & Deary, 2011). SNPs can lead to changes in the gene

product – either in function, production, or distribution. Candidate gene studies can provide insight into a relationship that is difficult to investigate in humans, but results should also be interpreted with caution since the effect size of one single gene on behavior is quite small. More recent investigations attempt to incorporate multiple gene variants or interactions with external factors (Raz & Lustig, 2014) and will be discussed further in the general discussion section.

For the purposes of this study we chose genes with SNPs that have a well-defined effect on components influencing the dopaminergic system (i.e. COMT, DAT1, DBH) and the levels of growth factors (i.e. BDNF and IGF1). Additionally, these neurochemicals are involved in cognition and are modulated with aging and variability in cardiorespiratory fitness level (see Table 1.1 for a review of the genes). Each gene will be discussed in detail in the following chapters.

Summary

The purpose of this thesis is to explore the potential interaction between genetics and cardiorespiratory fitness level. What contributes to the variability in the amount of beneficial effects that are seen with increases in cardiorespiratory fitness level? Could it be that there is a predisposition for the amount of benefit one can receive from increased cardiorespiratory fitness? Is it possible that our genetics can determine to what extent our environment will influence our brain functioning or cognitive abilities?

Or from a perpendicular perspective, is it possible that we can "rescue" ourselves from a predisposition for a disadvantaged cognitive ability? Is it possible that some of us have "bad" genes that determine that our attention spans or multitasking abilities are "bad"? But, if we exercise and increase our cardiorespiratory fitness levels, might we show greater cognitive benefits because we are starting out at a "lower" baseline than others? Whereas if we have the "good" genes, then there really isn't much room for us to improve, but being healthy is always better for a long and healthy life. The interaction between genetics and cardiorespiratory fitness could be a reason why we see inter-individual variability in the benefits of exercise interventions.

To test this potential interaction between genes and cardiorespiratory fitness level on cognition in older adults, we conducted a cross-sectional study with a group of healthy

sedentary older adults. We collected cardiorespiratory fitness levels and a complete neuropsychological battery of tests specifically chosen to investigate different aspects of cognition. We also collected blood to genotype particpants for a variety of candidate genes known to affect the dopamine system or neurotrophic factors. This thesis presents a statistical analysis of the effects of genotype alone and more importantly, of the interactive effect between genotype and cardiorespiratory fitness level on multiple behavioral measures in healthy sedentary older adults.

Table 1.1. Description of genes and SNPs used in this study

Dopamine-related Genes

Catechol-O-methyl Transferase (COMT) is an enzyme that breaks down dopamine (DA) and particularly important in the prefrontal cortex (PFC).

SNP	Effect	Genotypes	Enyme activity	DA level	Behav In Lit
COMT Val158Met (rs4680)					
G to A nucleotide switch causes a methionine (Met)	Changes thermostability	Val/Val	high	low	worst
to replace a valine (Val) at codon 158	of enzyme which affects the	Met/Val	med	med	
	level of activity and thus PFC	Met/Met	low	high	best
	dopamine levels				
COMT C/G (rs4818)					
C to G nucleotide mutation	Affects mRNA structure and	G/G	high	low	worst
	in turn protein expression	C/G	med	med	
		C/C	low	high	best

Dopamine Beta Hydroxylase (DBH) is an enzyme that converts dopamine into norepinephrine.

SNP	Effect	Genotypes	Enyme activity	DA level	Behav In Lit
DBH 444 G/A (rs1108580)					
G to A substitution at position 444,	Located at splice junction and	G/G	high	low	best
exon 2 on chromosome 9	may alter efficiency of mRNA	A/G	med	med	
	splicing therby affecting	A/A	low	high	worst
	levels of mature DBH mRNA				
DBH -1021 C/T (rs1611115)					
C to T substitution 1021 base pairs upstream	Affects transcription of the	C/C	high	low	best
of the transcriptional start site in the 5'-flanking	DBH gene	T/C	med	med	
region of the DBH gene		T/T	low	high	worst

Growth Factor Genes

Insulin-like Growth Factor I (IGF1) is a somatomedin that important for mammalian growth and development.

SNP	Effect	Genotypes	IGF1 serum level	Behav In Lit
IGF1 G/A (rs6220)				
G to A switch in exon 4 of 3' unstranslated region	Alters mRNA stability	A/A	low	worst
	and regulation of	A/G		
	protein expression	G/G	high	best

Brain-derived neurotrophic factor (BDNF) is a neurotrophin that contributes to brain health.

SNP	Effect	Genotypes	BDNF Secretion	Behav In Lit
BDNF Val66Met (rs6265)				
G to A switch at nucleotide 196 results in avaline (Val)	Affects activity dependent	Met/Met	low	worst
to methionine (Met) substitution at codon 66	secretion of BDNF	Met/Val		
		Val/Val	high	best

Chapter 2: General Methods

Participants

Older adults between the ages of 58 and 81 years of age were recruited from the local community of Urbana-Champaign, Illinois (N = 842). Blood was collected from 242 participants and sent to collaborators for genotyping. The final sample size used in this study varied depending on voluntary drop out, failure to complete the maximal graded exercise test, failure to complete or perform better than chance (e.g. < 50% error rate) on any test(s) in the neuropsychological test battery, failure to complete the first fMRI session, or a failure in genotyping for any of the genes of interest (COMT, DBH, DAT1, BNDF, IGF1). Sample sizes are reported in individual sections of the thesis.

Eligible participants had to demonstrate strong right handedness with 75% or above on the Edinburgh Handedness Questionnaire (Oldfield, 1971), score < 3 (total 15) on the Geriatric Depression Scale (Sheikh & Yesavage, 1986; Yesavage et al., 1982), have normal color vision and a corrected visual acuity of at least 20/40. All participants met or surpassed all criteria for participating in a magnetic resonance imaging study including no previous head trauma, no previous head or neck surgery, no diagnosis of diabetes, no neuropsychiatric or neurological condition including brain tumors, and no metallic implants that could interfere with or cause injury due to a magnetic field. Additionally, all participants received physician's clearance to engage in a maximal graded exercise test. Finally, all participants signed an informed consent approved by the University of Illinois and were compensated for their participation.

Blood Collection

Following an overnight fast, venous blood samples were collected into PAXgene DNA collection tubes (PreAnalytiX by Qiagen, Valencia, CA) and stored at -80°C until analysis. One tube containing ~10 ml of whole blood was sent per subject. All tubes were sent on dry ice to prevent thawing during shipment to collaborators at George Mason University where the genotyping took place. Confirmation that the samples arrived in satisfactory condition was made via e-mail.

DNA Preparation

DNA was purified from whole blood with a Flexigene DNA kit, following the procedure recommended by the manufacturer (Qiagen, Valencia, CA). DNA yields were quantified with a Nanodrop UV/visible spectrophotometer (Thermo Scientific, Wilmington, DE).

Genotyping

Genotyping was conducted at George Mason University and methods are described in detail within each chapter.

Physical Assessment

Cardiorespiratory fitness level. Participants were required to obtain consent from their personal physician before cardiorespiratory fitness testing was conducted. Cardiorespiratory fitness ($VO_{2\ max}$) was assessed by graded maximal exercise testing on a motor-driven treadmill. The protocol involved the participant walking at a speed slightly faster than normal walking pace (~30-100 m/min) with increasing grade increments of 2% every 2 minutes. A cardiologist and nurse continuously monitored measurements of oxygen uptake, heart rate, and blood pressure. Oxygen uptake (VO_2) was measured from expired air samples taken at 30-second intervals until a maximal VO_2 was attained or to the point of test termination due to symptom limitation and/or volitional exhaustion. $VO_{2\ max}$ was defined as the highest recorded VO_2 value when two of three criteria were satisfied: 1) a plateau in $VO_{2\ peak}$ between two or more workloads, 2) a respiratory exchange ratio > 1.00, and 3) a heart rate equivalent to their age predicted maximum (i.e. 220 – age). The primary measure was $VO_{2\ max}$ (ml/kg), with higher values implicating higher cardiorespiratory fitness levels. From here on VO_2 will be used interchangeably with $VO_{2\ max}$.

Other measures. $Weight_{kg}$, $height_m$, and body mass index (BMI; $weight_{kg}/height_m^2$) were also recorded.

Cognitive Assessment

Test administration was either computerized or paper-and-pencil. Response times were recorded either on a button box or keyboard with millisecond accuracy for tasks requiring response time (RT) measures. A stopwatch was used for timed paper-and-pencil tests. Participants were told to respond as quickly and as accurately as possible for timed tests.

Verbal crystallized intelligence test.

***Kaufman brief intelligence test – verbal subscale*.** The verbal subscale of the Kaufman Brief Intelligence Test (K-BIT; Kaufman & Kaufman, 1990) was administered to the participants by lab personnel. This subscale consists of verbal knowledge and riddles, both assessing crystallized verbal ability. The primary measure was an age-scaled score of correct items divided by total number of items attempted; therefore, higher scores indicate better performance.

Memory tests.

Digit span test. Two versions of the digit span test, forward and backward, were administered verbally. For the forward version, a task administrator read a sequence of numbers out loud to participants (e.g. 4-5-1) who were instructed to repeat the sequence of numbers back to the administrator in the exact order read (e.g. 4-5-1). For the backward version, the task administrator read a sequence of numbers out loud to participants (e.g. 4-5) who were instructed to repeat the sequence of numbers back to the administrator in reverse order (e.g. 5-4).

For both versions, participants were given two trials for each list length (e.g. 4-5-1, then 2-4-9). For the forward version, list lengths began with a three number sequence and incrementally increased by one number if participants could correctly repeat the sequence for at least one of the trials. If participants incorrectly repeated the sequences of a particular list length in both trials, the previous list length was recorded as the maximum digit span. The test was conducted in the same way for the backward version, but list lengths began with a two number sequence. The primary measure was the number of

digits in the maximum digit span (i.e. forward and backward separately); therefore, higher scores indicate better performance.

Spatial memory test. Participants performed a spatial memory paradigm that has been previously associated with cardiorespiratory fitness and hippocampal volume in a sample of 165 older adults (methods adapted from Erickson et al., 2009b). Participants were first presented with a fixation crosshair ('+') at the center of the screen for one second and instructed to keep their eyes on the crosshair. Following the fixation, 1, 2, or 3 black dots appeared at random locations on the screen for a duration of 500 ms. The dots were removed from the display and the fixation cross re-appeared on the screen for 3 seconds. During this time, participants were instructed to try and remember the location(s) of the previously presented black dot(s). At the end of the 3 s delay, a red probe dot appeared on the screen in either the same location as one of the target dots (match condition) or at a different location (non-match condition). Participants had 2 s to respond to the red dot by pressing the "M" key for a match trial or the "X" key for a non-match trial. Forty trials were presented for each set size (1, 2, or 3 locations), with 20 match trials and 20 non-match trials. Several practice trials were performed before the task began in order to acquaint the participants with the task instructions and responses. Both response time (RT) and error rate (ER) were recorded for each memory load and median RT and ER were used as the primary measure; therefore, lower scores indicate better performance.

Executive control function tests.

Verbal fluency test (FAS). Participants were asked to say as many words as they could, excluding proper nouns, in 60 seconds from a given phonetic category. Each participant had 60 seconds to name as many words as possible that began with the letter F, then 60 seconds for words beginning with A, and then 60 seconds for S. The primary measure was the total number of words named across categories; therefore, higher scores indicate better performance.

Flanker test. A modified Flanker paradigm (Botvinick, Nystrom, Fissell, Carter, & Cohen, 1999) was administered on a computer with a standard keyboard. For this task, participants were presented with five arrows in a row and instructed to indicate the orientation of the central arrow during two trial types, congruent and incongruent. Arrows

flanking the central arrow pointed in the same direction as the central arrow during congruent trials (e.g. >>>>>), and in the opposite direction during incongruent trials (e.g. >><>>). If the central arrow pointed to the left, participants were instructed to press the "Z" key, and if the central arrow pointed to the right, they were to press the "M" key. Both RT and ER were recorded for each trial type and median RT and ER were used as the primary measure. Cost scores were also calculated by subtracting the congruent condition median RT or ER from the incongruent condition median RT or ER; therefore, lower scores indicate better performance.

Stroop test. Behavioral data from a modified version of the color-word Stroop test (Erickson et al., 2009a; Milham et al., 2001) was collected within an fMRI environment. Participants were presented with congruent, neutral, incongruent-ineligible, and incongruent-eligible stimuli in an event-related design. Congruent stimuli were color words that matched the printed ink color (e.g. 'RED' in red ink). Neutral stimuli were words not associated with color that were matched for frequency and length with the color words (e.g. 'SHIP' in red ink). Incongruent-ineligible stimuli were color words that did not match the set of potential responses, and thus did not match the printed ink color (e.g. 'BLUE' in purple ink if 'blue' is not one of the potential responses). Incongruent-eligible stimuli were color words that did match the set of potential responses, but did not match the printed ink color (e.g. RED in purple ink and 'red' is one of the potential responses). There were two ink color sets that were counterbalanced across participants: 1) eligible = red, orange, and purple; ineligible = blue, green, and yellow, and 2) eligible = blue, green, and yellow; ineligible = red, orange, and purple. Participants responded with a simple button press on a button pad placed in their right hand with four buttons on the top (corresponding to each finger) and one button on the side (corresponding to the thumb). Depending on the eligible ink color set, the right index finger corresponded to red or blue, the right middle finger to orange or green, and the right ring finger to purple or yellow. Task administrators placed participants' fingers on the correct corresponding buttons.

Each condition consisted of 36 trials. For the incongruent-eligible condition, each ink color was paired with both response eligible color words 12 times during a session (6 times per word). However, for the incongruent-ineligible condition, each ink color was paired with one of the three ineligible words 12 times (4 times per word). Each stimulus

was displayed for 1 s with a 1.5 s response window and a 3 s stimulus-onset asynchrony (SOA). A fixation crosshair ('+') was presented on the screen during all interstimulus intervals. An event-related stimulus design with 40% jitter was employed, such that the timing between trials varied to optimize the stimulus sequence and timing. Stimulus sequence and jitter for each participant was generated using OptSeq2 (http://surfer.nmr.mgh.harvard.edu/optseq/). RT and ER were recorded for each trial type and median RT and ER were used as the primary measure.

Wisconsin card sorting test (WCST). Participants completed a computerized version of the WCST (Psychological Assessment Resources, Inc.). Participants were required to sort cards displayed on the computer screen. The cards contained geometric designs and could be sorted into categories by shape, color, or number of items. The participants were asked to match each card that appeared in the lower portion of the computer screen with one of the four cards displayed at the top of the screen. The participants were told that the program would provide feedback about the accuracy of their decision ('correct' or 'incorrect'), but the examiner could not give any additional instructions after the task began. The participant did not know the following, but after several trials of sorting cards correctly by a specific category, the category was changed arbitrarily, requiring the participant to adjust sorting accordingly. Total errors and percent perseverative errors were used as the primary measures; therefore, lower scores indicate better performance.

Task switch test. Participants were required to switch between judging whether a number (1, 2, 3, 4, 6, 7, 8, or 9) was odd or even and judging whether it was low or high (i.e. less than or greater than 5). Numbers were presented individually for 1500 ms against a pink or blue background at the center of the screen with the constraint that the same number did not appear twice in succession. If the background was blue, participants used one hand to report as quickly as possible whether the number was high ("X" key) or low ("Z" key). If the background was pink, participants used their other hand to report as quickly as possible whether the number was odd ("N" key) or even ("M" key). Participants completed four single task blocks (2 blocks odd/even and 2 blocks high/low; single condition) of 24 trials each. Due to the difficulty of this task, participants then completed a practice dual task block in which they switched from one task to the other for 120 trials.

Finally, they completed a dual task block of 120 trials during which the task for each trial was chosen randomly (mix condition). Within the mix condition, there were two trial types: 1) repeat trials in which the preceding trial involved the same rule set (e.g. odd/even trial followed by odd/even trial) and 2) switch trials were those when the preceding trial required the other rule set (e.g. odd/even trial followed by high/low trial). This task was similar to that of Kramer, Hahn, & Gopher (1999) and Pashler (2000); this version of the task was programmed and administered using E-prime software (Psychology Software Tools). Both RT and ER were recorded. The primary measures for this task were median RT and ER for each condition as well as each trial type within the mix condition. A global (mix condition – single condition) and a local (switch trial – repeat trial) cost score was also calculated for both RT and ER. Lower scores indicate better performance.

Dual task test. Behavioral data for this task was collected within an fMRI environment. Participants were given separate button pads for each hand with four buttons on the top (corresponding to each finger) and one button on the side (corresponding to the thumb). Task administrators placed participants' fingers on the correct corresponding buttons. During the task, participants were presented with two task conditions, single trials and dual trials, in an event-related design. During single task trials, participants were presented with either a single letter stimulus ('A' or 'B') or a single number stimulus ('2' or '3'). An asterisk was always presented above or below the single stimulus to act as a placeholder to match the dual task trial visual load. Instructions asked participants to indicate the identity of the stimuli with a simple button press. The left middle finger corresponded to the number '2', the left index finger to the number '3', the right index finger to the letter 'A', and the right middle finger to the letter 'B'. During dual task trials, participants were presented with both a letter and a number (one above the other), and instructed to indicate the identity of both stimuli with corresponding button presses. Participants were told that they could respond to either the letter or number first, but that buttons should never be pressed simultaneously. Each condition consisted of 48 trials (single task condition had 24 letter and 24 number trials). Each stimulus was displayed for 3 s with a 3 s response window and a stimulus onset asynchrony (SOA) of 1.5 s. A crosshair ('+') was presented on the screen during all interstimulus intervals. An event related stimulus design with a 40% jigger was employed, such that the timing between

trials varied in order to optimize the stimulus sequence and timing. Stimulus sequence and jitter for each participant was generated using OptSeq2 (http://surfer.nmr.mgh.harvard.edu/optseq/). Both RT and ER were recorded. For dual task trials, RTs for the first and second button press were recorded separately and a trial was counted as correct only if both the letter and number were identified correctly. The RT recorded from the second button press was used as the measure for dual trials. The primary measures were median RT and ER for each trial type. Cost scores were also calculated for both RT and ER by subtracting the single trial measure from the dual trial measure. Lower scores indicate better performance.

Statistical Methods

Exact tests were used to ensure that the genotypic distributions were consistent with Hardy-Weinberg equilibrium.

Analysis of variance and chi-square analyses were performed to determine whether genetic groups were equivalent with respect to potentially confounding variables (i.e. age, education, sex, mMMSE, BMI and VO_2).

The specific analyses are described in the specific sections. For figures representing genotypic analyses (supplementary figures in Appendix A and B), displayed values are estimated marginal means calculated after taking into account the control variables added into the model. Error bar values are the standard error values taken from bootstrap results based on 5000 bootstrap samples. For figures representing the interaction analyses (i.e. relationship between behavioral measure and cardiorespiratory fitness level for each genotypic/allelic group), all data points plotted in the scatter plot are raw, unadjusted scores, with lines fitted to the raw data for each group. The ΔR^2 and p value are also included, and if significant ($p \leq .05$) or marginal, ($p \leq .10$), β and p value for each group is also included.

Chapter 3: Dopamine Related Genes

Dopamine (DA) is best described as a modulatory neurotransmitter that is related to the gating of inputs and the modulation of postsynaptic neuronal activity through excitation or inhibition of neurons (Cooper, 2003, Chapter 9; Seamans & Yang, 2004). It plays an especially important modulatory role within the frontal cortex, which is critical for intact cognitive functioning (Bäckman, Nyberg, Lindenberger, Li, & Farde, 2006; Brozoski, Brown, Rosvold, & Goldman, 1979; Cools & Robbins, 2004; Savitz, Solms, & Ramesar, 2006; Seamans & Yang, 2004). Evidence for this was found in young adult monkeys who showed performance deficits on a spatial delayed alternation task after DA levels in the prefrontal cortex (PFC) were reduced by 80% through chemical injection of a selective catecholaminergic toxin, and these performance deficits were similar to those found in monkeys with ablated PFCs (Brozoski et al., 1979). Furthermore, reversal of these deficits was observed after administration of L-DOPA, a synthetic precursor for DA that can cross the blood brain barrier and subsequently be converted into DA. Beyond primate studies, in humans severe cognitive deficits are known to accompany Parkinson's Disease (PD) and Huntington's Disease (HD) in which the dopaminergic fronto-striatal system becomes severely impaired (Bäckman et al., 2006; Bäckman & Farde, 2001; Nieoullon, 2002).

The fact that DA plays a significant role in cognitive functioning is apparent, but the neurobiological mechanism behind the DA system is complex. Dopamine can exhibit opposing functional effects depending on the level of stimulation, the particular synapse, neuron, receptor, or level of activity within the local network being acted upon (Seamans & Yang, 2004). DA is believed to affect cognition in an inverted-U shaped function such that moderate levels are ideal while high and low levels can be detrimental (Cools & Robbins, 2004; Seamans & Yang, 2004).

Part of this complexity is inherent in the fact that in the central nervous system (CNS), DA interacts with at least five receptor subtypes (D1, D2, D3, D4, D5) that can be subdivided into two families. The D1-like family includes the D1 and D5 receptor subtypes, and the D2-like family includes the D2, D3, and D4 receptor subtypes (Cooper, 2003; Seamans & Yang, 2004). DA can exert both direct and indirect effects on the excitability of prefrontal neurons through activation of these different receptors. Both families of receptors can be found post-synaptically, but some D2-like receptors are also found pre-

synaptically where they act as autoreceptors (Bäckman et al., 2006; Cooper, 2003). Activation of these autoreceptors can slow firing rate and inhibit DA synthesis and release, thus they can be divided into 3 categories: 1) synthesis-modulating, 2) release-modulating, and 3) impulse-modulating (Cooper, 2003). It is interesting to note that the prefrontal and cingulate cortices lack synthesis-modulating autoreceptors, and thus can only regulate the release of DA and neuronal firing rates. D2 autoreceptors are 5-10 times more sensitive then post-synaptic receptors (Cooper, 2003), therefore low levels of DA will only stimulate autoreceptors, whereas high doses can stimulate both pre- and post-synaptic receptors.

In general, D1-like receptor activations lead to increased excitability while activation of D2-like receptors seem to suppress excitability (Seamans & Yang, 2004). Similar to DA levels, D1-like receptor stimulation has an inverted U-shaped effect on behavior with over stimulation or blockade of D1-like receptors in the prefrontal cortex leading to disruptions. For example, using a delayed spatial win-shift task, Floresco and Phillips (2001) found that an injection of D1 agonist in the rat prefrontal cortex before a test phase, but after a 30-minute delay period, disrupted performance. It was believed that there was an ideal amount of DA available to perform the task after the 30-minute delay, so the addition of D1 agonist overloaded the circuit. However, an injection of the D1 agonist after a 12-hour delay improved performance when information from the training phase was degrading and the addition of the D1 agonist was able to boost the circuit (Floresco & Phillips, 2001).

In humans, molecular imaging studies show that DA release in the frontal cortex and hippocampus is related to working memory performance (Aalto, Bruck, Laine, Nagren, & Rinne, 2005) while striatal DA release is seen during card-sorting (Monchi, Hyun Ko, & Strafella, 2006) and sequential learning (Badgaiyan, Fischman, & Alpert, 2007) in young adults. DA markers are also strongly related to performance in executively demanding tasks (Bäckman et al., 2000; Erixon-Lindroth et al., 2005; Volkow et al., 1998), which show the most pronounced age-related deficits (Winter et al., 2007). Taken together, this indicates that age-related differences observed in executively demanding tasks are partially a reflection of age-related deficits of the DA system and its ability to meet performance demands (Mattay et al., 2003).

The process of aging brings about general neurochemical changes in the brain such as region-specific declines in the concentration, synthesis, and number of receptor sites of neurotransmitters such as serotonin, acetylcholine, and dopamine. Small alterations in dopaminergic fronto-striatal loops may be an important contributor to the cognitive decline in normal aging due to the dense innervation between the caudate nucleus and the frontal cortex (Cabeza, 2001a). Mirroring volume losses, receptor losses are most conspicuous in the prefrontal cortex (Cabeza, 2001a; Strong, 1998). Goldman-Rakic and Brown (1981) observed a drastic 56% reduction in the concentration of dopamine (DA) in the prefrontal cortex of old monkeys (18 yrs) compared to young monkeys (2-3 yrs), while the levels of norepinephrine (NE) and serotonin (5-HT) levels remained relatively stable. DA was also reduced in the superior and posterior inferotemporal cortex and caudate nucleus by an average of 39%, 35% and 34% respectively. In contrast, levels in the parietal and occipital cortices and the hippocampus were unchanged across age. Not only was there a reduction in the concentration of DA, but catecholamine biosynthesis was also reduced by more than 60% in the prefrontal, temporal, parietal and occipital cortices. Furthermore, these neurochemical changes also translate into behavioral changes. Bartus, Flemin, & Johnson (1978) found that ≥18 yr old rhesus monkeys were severely impaired on a spatial delayed alternation task, the same task on which young adult monkeys were impaired after injection of a catecholaminergic toxin mentioned earlier (Brozoski et al., 1979).

In a recent review of human literature, Li & Rieckmann (2014) summarized that pre- and post-synaptic dopamine receptor density levels can decline up to 10% per decade, beginning around the third decade of life, and these changes in the dopamine system can translate to changes in cognition. For example, in a recent review, Ranganath & Jacob (2015) discussed studies that showed the importance of prefrontal D1 and D2 receptors in cognitive flexibility in non-human animals, concluding that D1 receptors may be necessary for stabilizing new mental representations after identification of effective strategies, while D2 receptors are important for the destabilization of prefrontal networks, allowing the exploration of new strategies. Part of the translation from dopamine to behavior stems from the key role dopamine plays in the tuning of the signal-noise-ratio (SNR) of neuronal signal transmission, known as neuronal gain control. Neurocomputational models have

provided insight into the behavioral effects caused by alterations in the SNR. For example, a simulated "old" network shows that reducing gain control leads to a reduced slope of the activation function and the SNR of information transmission results in suboptimal effects to neurocomputation. These effects include increases in random processing activations (i.e. noise) and reduction in the representation distinctiveness of activation patterns, which is similar to neuroimaging studies that see more diffuse brain activation patterns in older adults compared to more distinct patterns seen in young adults while performing the same task (Li & Rieckmann, 2014; Sikström, 2007). Alterations in the gain parameter also mirrors the inverted U-shaped curve of behavioral performance as a function of dopamine levels, providing more insight into the relationship between dopamine and behavior and the need for a delicate balance.

In contrast to aging, exercise has been associated with increases in levels of various neurochemicals and improvements in overall brain health. For example, regular physical exercise in rats, specifically wheel-running, increases levels of brain catecholamines, particularly dopamine and noradrenaline (Sutoo & Akiyama, 2003). Further, exercise-induced increases in levels of brain-derived neurotrophic factor lead to increased neuronal survival (Berchtold, Castello, & Cotman, 2010; Vaynman & Gomez-Pinilla, 2006) and even neurogenesis (Creer, Romberg, Saksida, van Praag, & Bussey, 2010; van Praag, Kempermann, et al., 1999). Further, exercise does not only induce short-term increases in neurotransmitter levels, but Meeusen et al. (1997) also showed that exercise training produced specific adaptations in basal neurotransmitter output in the rat striatum.

In human studies, peripheral measures of dopamine are often used as indicators of central levels under the assumption that brain and systemic DA levels respond similarly to physical exercise. For example, Winter et al. (2007) found a positive linear correlation of elevated levels of peripheral DA and epinephrine with better retention of learned verbal material, and concluded that DA and epinephrine are mediators through which physical exercise improves learning.

Since exercise can increase cardiorespiratory fitness levels, variations in cardiorespiratory fitness level may lead to individual variations in the amount and availability of dopamine. If this holds true, there is a possibility that behavioral

performance can be influenced by an interaction between cardiorespiratory fitness level and genes that specifically affect dopamine.

Methods

Sample

The initial sample size of *N* = 242 was reduced due to participant drop out, experimenter error, failure to complete behavioral or fitness testing, and genotyping failure. The sample size was then reduced further with pairwise deletion of participants that performed below chance (i.e. > 50% error rate) on any behavioral test. Therefore, the sample size for each test varies and each is indicated within the results section.

Genotyping

DBH 444 G/A (rs1108580). The DBH 444 G/A polymorphism was assayed after nested PCR, by using allele-specific T_m-shift primers in various combinations, together with automated melting curve analysis as previously described (Greenwood et al., 2009). The yields of the PCR products corresponding to the A and G alleles were equalized by adjusting the concentrations of their respective oligonucleotide primers.

DBH –1021 C/T (rs1611115). The DBH -1021 C/T polymorphism was assayed after nested PCR, by using allele-specific T_m-shift primers in various combinations, together with automated melting curve analysis (Lipsky et al., 2001; Wang, Kong, Zhang, Sun, & Geller, 2005) on an iCycler real-time PCR machine (Bio-Rad, Hercules, CA). The PCR product of the first round (outer) amplification was 344 base pairs (bp) in length. The PCR product of the second round (inner) amplification was 106 bp for the C allele, and 92 bp for the T allele.

COMT Val158Met (rs4680) and COMT C/G (rs4818). The COMT Val158Met and COMT C/G polymorphisms were assayed by DNA sequencing. First, a PCR fragment 290 base pairs in length was amplified by PCR and purified by ethanol precipitation. Then the DNA sequence of this PCR fragment was determined by cycle sequencing with BigDye terminators on an ABI 310 capillary sequencer (Applied Biosystems, Carlsbad, CA). The Fryxell lab and other labs have previously shown that this approach allows reliable

determination of both homozygous and heterozygous SNP genotypes (Hare & Palumbi, 1999; Söderlund, Canto, & Méndez, 2002).

Statistical Analyses

Hardy-Weinberg equilibrium. Two hundred and forty-two samples were genotyped successfully for the DBH gene. The frequencies of the three DBH 444 G/A genotypes were .19 for G/G, .54 for G/A, and .27 for A/A. The frequencies of the three DBH -1021 C/T genotypes were .71 for C/C, .27 for CT, and .02 for TT. The Hardy-Weinberg exact test showed the allelic distributions for both SNPs did not deviate from those expected, DBH 444 G/A: $\chi^2(1) = 1.87$, $p = .197$; DBH -1021 C/T: $\chi^2(1) = 0.00$, $p = 1.00$.

Two hundred and forty-one samples were genotyped successfully for the COMT gene. The frequencies of the three COMT Val158Met genotypes were .17 for G/G (Val/Val), .54 for G/A (Val/Met), and .29 (Met/Met). The frequencies for the COMT C/G genotypes were .11 for G/G, .48 for GC, and .41 for C/C. The Hardy-Weinberg exact test showed the allelic distributions for both SNPs did not deviate from expectations, COMT Val158Met: $\chi^2(1) = 2.51$, $p = .150$; COMT C/G: $\chi^2(1) = 0.55$, $p = .571$.

SNP analysis. All performance measures were analyzed using analysis of covariance (ANCOVA) with age, education, and sex as covariates. Percentile bootstrapped 95% confidence intervals (CI) were calculated over 5000 samples for pairwise comparisons and are reported in text if there was a significant omnibus effect of the SNP.

SNP x Fitness Level interaction analysis. All performance measures were analyzed using stepwise multiple regression with control variables entered in the first step (e.g. age, education, and sex). The dummy coded SNP variable(s) and cardiorespiratory fitness level (i.e. VO_2 scores) were both entered in the second step, and the SNP x Fitness Level interaction variable(s) was entered in the last step. If the SNP x Fitness Level interaction was significant (i.e. ΔR^2 was $p \leq .05$) or marginal (i.e. ΔR^2 was $p \leq .10$), simple slopes analysis was conducted to elucidate the nature of the interaction. For the simple slopes analysis, the control variables were entered in the first step, followed by the SNP dummy variable(s) in the second step, and then in the last step cardiorespiratory fitness level was added for each individual group (e.g. for DBH -1021 C/T, VO_2 scores for T Carriers

with C/C Homozygotes coded 0 and VO_2 scores for C/C Homozygotes with T Carriers coded 0 as two separate interaction variables). For each group, the unstandardized beta (*B*), standardized beta (β), *t* value, along with percentile bootstrapped *p* value and 95% confidence intervals (CI) calculated over 5000 samples are reported in text.

Cognitive Assessment

This section is included here to help remind the reader of the variables of interest for each particular test. Full descriptions of the tests can be found in Chapter 2: General Methods.

Verbal crystallized intelligence. One test was used to assess verbal crystallized intelligence.

K-BIT test. The primary measure was an age-scaled score of correct items divided by the total number of items attempted on the verbal subscale of the Kaufman Brief Intelligence Test (K-BIT; Kaufman & Kaufman, 1990). Higher scores indicate better performance.

Memory. Two tests were used to assess various aspects of memory (i.e. short-term, working, and spatial).

Digit span test. Two versions of the digit span test, forward and backward, were used to assess short-term and working memory, respectively. For both versions of the digit span, the primary measure was the total number of digits in the maximum span length repeated; therefore, higher scores indicate better performance.

Spatial memory test. The primary measures were response time (RT) and error rate (ER) for each of the three memory loads (i.e. 1, 2 or 3). Lower scores (i.e. faster RT and lower ER) indicate better performance.

Executive control functioning. Six tests were used to assess various aspects of executive control functioning.

Verbal fluency test (FAS). The verbal fluency test assessed fluid intelligence and the primary measure was the total number of words named in 180 seconds for three phonetic categories (i.e. 60 seconds for F, A, & S separately). Higher scores indicate better performance.

Flanker test. The Flanker test assessed inhibitory processing. The primary measures were RT and ER for the two trial types (i.e. congruent and incongruent). Comparison of these two trial types assessed the ability to inhibit conflicting information. Lower scores indicate better performance.

Wisconsin card sorting test. The Wisconsin Card Sorting Test (WCST) assessed many aspects of executive functioning including inhibitory processing and concept shifting. The primary measures were percent perseverative errors and total errors; therefore, lower scores indicate better performance.

Task switch test. The task switch test assessed maintenance and coordination of multiple sets. The primary measures were RT and ER for the two conditions (i.e. single and mix), which can be compared to assess the cost of maintaining one versus two rule sets. The mix condition can be further split into repeat and switch trial types, which can be compared to assess the cost of rule switching. Lower scores indicate better performance.

Dual task test. The dual task test assessed coordination of multiple sets. The primary measures were RT and ER for the two trial types (i.e. single and dual), which can be compared to assess the cost of responding to one versus two stimuli. Lower scores indicate better performance.

Dopamine Beta Hydroxylase

Dopamine beta hydroxylase (DBH) is an enzyme that catalyzes the conversion of dopamine (DA) to norepinephrine (NE; Cubells et al., 1998; Greene, Braet, Johnson, & Bellgrove, 2008; Parasuraman et al., 2005). Activity of the enzyme is highly correlated with levels of the protein, thus differences in protein level are assumed to reflect differences in enzyme activity (Cubells & Zabetian, 2004; Cubells et al., 1998). DBH found in plasma arises from the sympathetic nervous system (Cubells et al., 1998), while DBH levels in cerebrospinal fluid (CSF) are controlled by central noradrenergic neurons, where DBH is localized within synaptic vesicles and released together with NE (Cubells & Zabetian, 2004). The locus ceruleus contains the largest population of noradrenergic neurons and accounts for most of the noradrenergic innervation of the forebrain (Cubells & Zabetian, 2004). Although plasma and CSF DBH are under the control of different systems, a strong

correlation has been found between plasma DBH activity and CSF DBH activity (Hess et al., 2009).

Levels of DBH are under strong genetic control by the DBH gene such that heritability estimates are .98 for plasma levels and .83 for CSF (Hess et al., 2009). The DBH gene is known to have several polymorphisms that are in strong linkage disequilibrium (Kopecková, Paclt, & Goetz, 2006). Linkage disequilibrium is defined as two or more alleles at different loci that occur together within an individual at a greater frequency than chance (Bellgrove & Mattingley, 2008). One of these single nucleotide polymorphisms (SNP) is a guanine (G) to adenine (A) substitution at position 444, exon 2 (DBH 444 G/A; rs1108580) on chromosome 9 (Cubells et al., 1998; Parasuraman et al., 2005). The A allele is associated with lower plasma and CSF levels of DBH, resulting in less enzymatic activity (i.e. higher DA:NE ratio; Cubells et al., 1998). This SNP does not alter the primary structure of the DBH protein, but it is located at the splice junction between exon 2 and intron 2 of the DBH gene, which may alter the efficiency of mRNA splicing, thereby affecting levels of the mature DBH mRNA (Cubells et al., 1998).

Another SNP of interest is a cytosine (C) to thymine (T) substitution 1021 base pairs upstream of the transcriptional start site in the 5'-flanking region of the DBH gene (DBH -1021 C/T; rs1611115) that is hypothesized to affect DBH gene transcription (Cubells & Zabetian, 2004; Zabetian et al., 2001). This SNP accounts for 35-52% of the total variance of DBH plasma activity (Hess et al., 2009; Zabetian et al., 2001). The T allele of the DBH -1021 C/T SNP is associated with low DBH plasma activity, while the C/C genotype shows the highest DBH activity (i.e. high DA:NE and low DA:NE ratio respectively; Hess et al., 2009). The T allele occurs at a frequency of approximately .22 in European populations, a rate considerably less frequent compared to the C allele (Zabetian et al., 2001). In 2003, Zabetian et al. showed that the alleles for each SNP occur together at the following frequencies: AC = .296, AT = .205, and CG = .429.

Although there is no direct research on the effect of either the DBH 444 G/A or DBH -1021 C/T SNPs on catecholamine levels in the human brain, a study on DBH knockout mice showed that the gene directly affects the balance of catecholamines in the prefrontal cortex (PFC; Bourdélat-Parks et al., 2005). Results indicated that norepinephrine (NE) was absent in the PFC of -/- mice, but only slightly reduced in +/- animals compared to wild-

type (+/+) animals. On the other hand, dopamine (DA) in the PFC was increased twofold in +/- mice, fivefold in -/- mice, and the NE to DA ratio was reduced by ~35% in +/- mice compared to wild-type mice. It is therefore postulated that DBH can act as a rate-limiting factor for NE synthesis, leading to less efficient conversion of DA to NE and elevating vesicular and synaptic DA:NE ratios (Cubells & Zabetian, 2004). This can be functionally important to regions that receive noradrenergic innervation and contain DA receptors, such as the PFC.

DBH Predictions

In terms of how polymorphisms in the DBH gene may affect cognition, Parasuraman et al. (2005) found that increasing gene dose of the G allele of the DBH 444 G/A SNP was associated with increased memory accuracy at the highest memory load (i.e. memory load of 3 dots) in a spatial memory task in middle-aged adults. This task required participants to remember the location(s) of 1, 2, or 3 dots over a short delay period and then indicate whether the location of a probe dot matched the location of one of the previously seen dots. This task required participants to update dot locations for each trial. Further, Greenwood et al. (2009) also found that G allele carriers in a mixed sample of young and old adults showed the best performance on a similar task.

In regards to the DBH -1021 C/T SNP, Greene et al. (2009) showed a linear decrease in errors of commission in a Sustained Attention to Response Task (SART) for increasing C allele dosage in healthy middle-aged individuals. The SART is an attentional task that requires participants to respond continuously to a stream of single digits (1-9), and to withhold response to a low-frequency digit (3) occurring only 11% of the time. A button press when the low-frequency digit appears is considered an error of commission indicating a lack of response inhibition. Therefore, increasing C allele dosage was associated with increased inhibitory processing (i.e. better executive control).

In both studies, better executive functioning was associated with the allele leading to increased DBH activity and a decrease in the DA/NE ratio translating to lower PFC DA levels. This seems counterintuitive to the extant literature indicating the importance of DA to executive functioning (e.g. Robbins, 2000; Cools & Robbins, 2004; Li, Lindenberger, &

Bäckman, 2010). However, it may be indicative of the delicate balance that needs to be maintained between the different neurotransmitters in the brain, such that more is not always better. With the severe age-related decline in DA and relative sparing of NE levels specific to the PFC (Goldman-Rakic & Brown, 1981), there may be a switch in the direction of the DBH genotypic effect across the lifespan. Therefore I predict that in the current study with a healthy older adult cohort, the alleles that translate to a higher PFC DA/NE ratio (i.e. A allele for DBH 444 G/A and T allele for DBH -1021 C/T) will be beneficial to both memory and executive processing (e.g. set switching and response inhibition) and thus show the best behavioral performance.

In regards to an interaction between the DBH gene x Fitness Level, I predict cardiorespiratory fitness level will have a stronger relationship to performance in carriers of the alleles that lead to a lower PFC DA/NE ratio (i.e. G allele for DBH 444 G/A and C allele for DBH -1021 C/T). This prediction is based on evidence that DA levels are increased by exercise and exercise can increase an individual's fitness level. Therefore, carriers of the alleles leading to a lower PFC DA/NE ratio have more to gain from increasing cardiorespiratory fitness levels.

DBH Results

DBH 444 G/A (rs1108580)

There were no significant differences between the genotypic groups on age, mMMSE, BMI or VO_2 (Table 3.1). There was a marginal difference between the groups on education, but this should not affect the results since education was controlled for in all analyses. Chi-square analysis showed there were no differences in the distribution of males and females between the groups. Means and standard deviations for the primary measures on the behavioral tests are reported in Table 3.2.

Verbal crystallized intelligence. Verbal crystallized intelligence is believed to remain relatively stable among older adults (Hedden & Gabrieli, 2004; Park & Gutchess, 2002). Therefore, I hypothesize that there will be no significant group differences on performance of the KBIT test, a measure of verbal crystallized intelligence. Further, I do not

expect the DBH 444 G/A x Fitness Level interaction to affect performance on the test of verbal crystallized intelligence.

KBIT test. As predicted, there was no effect of the DBH 444 G/A SNP or interaction between DBH 444 G/A X Fitness Level on performance for the KBIT test. The sample size for the KBIT test was $N = 194$, with genotypic group sizes as follows: G/G = 40, G/A = 106, A/A = 48. Analysis of covariance (ANCOVA) showed no significant differences between the groups, $F(2, 188) = 1.74$, $p = .178$.

When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 159$, with genotypic group sizes as follows: G/G = 35, G/A = 89, A/A = 35. Hierarchical regression showed that the interaction between DBH 444 G/A x Fitness Level did not significantly explain any variance on the age-scaled scores, $\Delta R^2 = .007$, $\Delta F(2, 150) = 0.732$, $p = .483$.

Memory tests. Performance on working and spatial memory tests decrease in older adults, in contrast to general sparing in tests of short term memory (Hedden & Gabrieli, 2004; Park & Gutchess, 2002). Previous literature in middle aged adults points to the G allele of the DBH 444 G/A SNP leading to better performance on tests of working and spatial memory (Greenwood et al., 2009; Parasuraman et al., 2005). However, I predict that in older adults, the A allele that leads to a higher DA:NE ratio will lead to better performance. Further, I expect an interaction between DBH 444 G/A and cardiorespiratory fitness level such that the G carriers (i.e. allele that leads to higher DA:NE ratio in animals; Cubells et al., 1998) will specifically show a positive relationship between behavioral performance and fitness level.

Digit span test. The sample size for the digit span test was $N = 196$ with group sizes as follows: G/G = 40, G/A = 107, A/A = 49. There were no significant group differences on either version of the test: forward span, $F(2, 190) < 1$; backward span, $F(2, 190) = 1.37$, $p = .257$. When cardiorespiratory fitness level was added to the model, the sample size was reduced to $N = 161$ with group sizes as follows: G/G = 35, G/A = 90, A/A = 36. The interaction between DBH 444 G/A x Fitness Level did not significantly explain variance in the length of span for either forward span, $\Delta R^2 = .008$, $\Delta F(2, 152) = 0.691$, $p = .503$, or backward span, $\Delta R^2 = .003$, $\Delta F(2, 152) = 0.233$, $p = .793$.

Spatial memory test. This study did not find an effect of the DBH 444 G/A SNP on a spatial memory test as previously reported in Greenwood et al., (2009; *N* = 96) and Parasuraman et al. (2005; *N* = 103). The sample size for the spatial memory test was *N* = 196 with genotypic group sizes as follows: G/G = 40, G/A = 107, A/A = 49. There were no significant differences between the genotypic groups for any memory load on either response time (RT) or error rate (ER; Table A.1).

When cardiorespiratory fitness level was added into the model, the sample size was reduced to *N* = 161 with genotypic group sizes as follows: G/G = 30, G/A = 90, A/A = 36. There was no significant interaction between DBH 444 G/A x Fitness Level on either RT or ER for any memory load of the spatial memory test (Table 3.3).

Executive control function tests. Aging is thought to have the greatest negative effect on performance of tests of executive control (Hedden & Gabrieli, 2004; Park & Gutchess, 2002) and age-related declines in the dopamine system are believed to contribute to this cognitive decline (Li & Rieckmann, 2014). Based on evidence that the A allele of the DBH 444 G/A SNP leads to a higher DA:NE ratio (Cubells et al., 1998), I predict carriers of the A allele will show better behavioral performance, while G allele carriers will specifically show a postitve relationship between behavioral performance and cardiorespiratory fitness levels.

Task switch test. The sample size for the task switch test was *N* = 157, with genotypic group sizes as follows: G/G = 30, G/A = 90, A/A = 37. Analysis of covariance (ANCOVA) showed no significant differences between the genotypic groups on response time (RT) for either condition: single condition, $F(2, 151) = 2.33$, $p = .101$; mix condition, $F(2, 151) = 1.06$, $p = .350$. However, there was a significant effect of genotype on error rate (ER) for the single condition, $F(2, 151) = 4.28$, $p = .016$ (Figure A.1a), with bootstrapped pairwise comparisons revealing the G/A group had a marginally lower ER compared to the G/G group, mean difference = -.040, $p = .058$, 95% CI [-.084, -.002], and the A/A group, mean difference = -.039, $p = .060$, 95% CI [-.080, -.002]. There was also a marginal genotypic effect on ER in the mix condition, $F(2, 151) = 2.79$, $p = .064$, with the G/G group having marginally more errors compared to the G/A group, mean difference = .048, $p = .083$, 95% CI [.003, .092], and the A/A group, mean difference = .057, $p = .069$, 95% CI [.005,

.109]. Further, there was a significant difference between the groups on a global error cost score (i.e. mix condition ER – single condition ER), $F(2, 151) = 3.34$, $p = .038$ (Figure A.1b), with the A/A group showing a significantly lower cost compared to both the G/A group, mean difference = -.048, $p = .031$, 95% CI [-.094, -.004], and marginally lower than the G/G group, mean difference = -.056, $p = .065$, 95% CI [-0.12, 0.00].

Further analysis of the mix condition showed no effect of the DBH 444 G/A SNP on RT for either repeat, $F(2, 151) = 2.13$, $p = .123$, or switch trials, $F(2, 151) < 1$, but there was a marginal genotypic effect on ER for repeat trials, $F(2, 151) = 2.60$, $p = .077$. Bootstrapped pairwise comparisons showed the G/G group had marginally higher ERs compared to both the G/A group, mean difference = .044, $p = .100$, 95% CI [-.005, .098], and the A/A group, mean difference = .051, $p = .089$, 95% CI [-.007, .111] (Figure A.2). There was also a marginal effect on ER for switch trials, $F(2, 151) = 2.61$, $p = .077$, with the G/G group again showing marginally higher ERs compared to the other groups: G/G vs G/A, mean difference = .051, $p = .094$, 95% CI [-.006, .114]; G/G vs A/A, mean difference = .063, $p = .066$, 95% CI [-.002, .132] (Figure A.2). However, there was no significant effect on a local error cost (i.e. switch ER – repeat ER), $F(2, 151) < 1$.

These results suggest that the DBH 444 G/A genotype may influence performance at a global level, maintenance of two sets as opposed to switching between the two tasks sets. These results also follow the prediction I made that in older adults the A allele leads to better performance in contrast to the reported results in middle aged adults where the G/G group showed better performance.

When cardiorespiratory fitness level was added into the model, the sample size for the task switch test was reduced to $N = 131$, with genotypic group sizes as follows: G/G = 26, G/A = 77, A/A = 28. There was no interaction between DBH 444 G/A x Fitness Level on response time (RT) for either condition: single condition, $\Delta R^2 = .017$, $\Delta F(2, 122) = 1.15$, $p = .320$; mix condition, $\Delta R^2 = .024$, $\Delta F(2, 122) = 1.72$, $p = .183$. For error rate (ER), there was no significant interaction effect for the single condition, $\Delta R^2 = .002$, $\Delta F(2, 122) < 1$ (Figure 3.1a), but the interaction did marginally explain 3.9% of the variance in ER for the mix condition, $\Delta R^2 = .039$, $\Delta F(2, 122) = 2.82$, $p = .063$. Simple slopes analysis showed ER significantly decreased across the mix condition trial types with increasing VO_2 for both the G/G group, $B = -0.015$, $\beta = -1.19$, $t(122) = -3.25$, $p = -.008$, 95% CI [-0.027, -0.003], and the

A/A group, B = -0.009, β = -0.73, $t(122)$ = -2.02, p = .038, 95% CI [-0.018, -0.001], but not the G/A group, B = -0.003, β = -0.37, $t(122)$ = -1.17, p = .188, 95% CI [-0.008, 0.002] (Figure 3.1b). However, there was no significant effect on a global error cost score (i.e. mix condition ER – single condition ER), ΔR^2 = .024, $\Delta F(2, 122)$ = 1.61, p = .205.

Further analysis of the task switch test mix condition showed no interaction effect on RT for either repeat, ΔR^2 = .018, $\Delta F(2, 122)$ = 1.26, p = .288, or switch trials, ΔR^2 = .021, $\Delta F(2, 122)$ = 1.48, p = .232. There was also no interaction effect on ER for repeat trials, ΔR^2 = .019, $\Delta F(2, 122)$ = 1.34, p = .266 (Figure 3.2a), but there was a significant effect on ER for switch trials, ΔR^2 = .057, $\Delta F(2, 122)$ = 4.308, p = .016. Simple slopes analysis showed a significant decrease in ER with increasing cardiorespiratory fitness level for the G/G group, B = -0.020, β = -1.41, $t(122)$ = -3.93, p = .001, 95% CI[-0.032, -0.009], marginally for the A/A group, B = -0.009, β = -0.68, $t(122)$ = -1.91, p = .057, 95% CI[-0.020, 0.000], but a non-significant relationship in the G/A group, B = -0.004, β = -0.38, $t(122)$ = -1.21, p = .175, 95% CI[-0.009, 0.002] (Figure 3.2b). However, there was no interaction effect on a local error cost (i.e. switch trial ER – repeat trial ER), ΔR^2 = .022, $\Delta F(2, 148)$ = 1.76, p = .176. This suggests that cardiorespiratory fitness level may not affect the groups equally.

DBH -1021 C/T (rs1611115)

There were no significant differences between the T Carriers and C/C Homozygotes on age, BMI or VO_2, however the groups did significantly differ on education and mMMSE (Table 3.4). Chi-square analysis revealed a significantly different distribution of males and females in the groups. For the C/C Homozygotes, there were more females and fewer males than expected, while the opposite was true for the T Carriers. However, these differences should not affect the results since sex and education were already planned to be used as control variables. Due to the difference found in mMMSE, it was also added as a control variable for the following analyses. Means and standard deviations for the primary measures on the behavioral tests are reported in Table 3.5.

Verbal crystallized intelligence. Verbal crystallized intelligence remains relatively stable in older adult populations (Hedden & Gabrieli, 2004). I do not expect to find a

significant allelic or interaction effect on performance of the measure of verbal crystallized intelligence.

KBIT test. The sample size for the KBIT test was $N = 194$, with allelic group sizes as follows: C/C Homozygotes = 136, T Carriers = 58. As expected, there were no significant differences between the allelic groups on the KBIT test, $F(1, 188) < 1$. When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 159$ with allelic group sizes: C/C Homozygotes = 109, T Carriers = 50. There was no significant interaction effect on the age-scaled scores, $\Delta R^2 = .003$, $\Delta F(1, 151) < 1$.

Memory tests. Performance on working and spatial memory tests decrease in older adults, in contrast to general sparing in tests of short term memory (Hedden & Gabrieli, 2004; Park et al., 2002). Further, the literature points to the DBH -1021 C/T SNP influencing both working and spatial memory, but not short term memory and I expect to find similar results in this study. Specifically, no allelic effect on a test of short-term memory (i.e. forward span), but that T Carriers will out perform the C/C Homozygotes on the tests of working (i.e. backward span) and spatial memory due to more availability of prefrontal dopamine. I also hypothesize an interaction effect on both the working and spatial memory tests such that the C/C Homozygotes will show a larger fitness effect, with no effect on the test of short term memory.

Digit span test. The sample size for the digit span test was $N = 196$, with allelic group sizes as follows: C/C Homozygotes = 138, T Carriers = 58. The allelic groups showed no significant differences on the number of digits for the forward span, $F(1, 190) < 1$ or the backward span, $F(1, 190) = 2.61$, $p = .108$.

When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 161$ with allelic groups as follows: C/C Homozygotes = 111, T Carriers = 50. The DBH -1021 C/T x Fitness Level interaction did not significantly explain any variance in forward span, $\Delta R^2 = .009$, $\Delta F(1, 153) = 1.59$, $p = .210$ (Figure 3.3a), but the interaction did significantly explain 2.9% of the variance in backward span, $\Delta R^2 = .029$, $\Delta F(1, 153) = 6.00$, $p = .015$. Although, simple slopes analysis showed a non-significant relationship between the length of span and cardiorespiratory fitness level for both C/C Homozygotes, $B = -0.03$, $\beta = -0.19$, $t(15) = -0.86$, $p = .396$, 95% CI [-0.08, 0.04] and T Carriers, $B = 0.09$, $\beta = 0.67$, $t(153) =$

2.00, p = .103, 95% CI [-0.01, 0.20], the slopes were significantly different from each other, B = 0.12, β = 0.86, $t(153)$ = 2.45, p = .032, 95% CI [0.01, 0.22] (Figure 3.3b). This suggests the relationship between working memory and cardiorespiratory fitness level differs for T Carriers and C/C Homozygotes.

Spatial memory test. Furthermore, T Carriers showed faster response times (RT) across all memory loads (i.e. 1, 2, and 3) on the spatial memory test, significant for memory load 1 RT, $F(1, 170)$ = 5.14, p = .025, mean difference = -65.43, p = .029, 95% CI [-122.79, -6.16] and memory load 2 RT, $F(1, 170)$ = 4.94, p = .028, mean difference = -62.50, p = .027, 95% CI [-116.79, -7.09], marginal for memory load 3 RT, $F(1, 170)$ = 2.34, p = .088, mean difference = -52.98, p = .083, 95% CI [-112.40, 6.54] (Figure A.3). The sample size was N = 176 with allelic group sizes as follows: C/C Homozygotes = 122, T Carriers = 54. Further, there were no significant differences on error rate (ER) for any memory load suggesting the differences in RT were not due to speed accuracy tradeoffs: memory load 1, $F(1, 170)$ = 1.36, p = .245; memory Load 2, $F(1, 170)$ = 1.34, p = .249; memory load 3, $F(1, 170) < 1$.

When cardiorespiratory fitness level was added into the model, the sample size was reduced to N = 144 with allelic group sizes as follows: C/C Homozygotes = 98, T Carriers = 46. The interaction between DBH -1021 C/T x Fitness Level had no significant effect on RT or ER for any memory load (Table 3.6).

Taken together, these results suggest the DBH -1021 C/T SNP influences working and spatial memory, but not short term memory. Further, the -1021 C/T SNP moderates the relationship between cardiorespiratory fitness level and spatial memory.

Executive control function tests. Aging is thought to have the greatest negative effect on performance of tests of executive control (Hedden & Gabrieli, 2004; Park & Gutchess, 2002). C/C Homozygotes have less DBH activity, implying a lower DA:NE ratio (Hess et al., 2009), and have been shown to perform worse on some measures of executive control (Greene et al., 2009). Therefore, I expect to find the same allelic effect (i.e. C/C Homozygotes show poor performance) on the following tests of executive control function. Further, higher levels of fitness are associated with better performance on measures of executive control (Colcombe et al., 2004a). Taken together, I expect to find an interaction

effect such that the C/C Homozygotes will show a benefit from increasing cardiorespiratory fitness level, but the T Carriers will not show this relationship.

Flanker test. For the Flanker test, the sample size was N = 187 with allelic group sizes as follows: C/C Homozygotes = 132, T Carriers = 55. The T Carriers were significantly faster than C/C Homozygotes on both trial types: congruent, $F(1, 181) = 4.13$, $p = .044$, mean difference = -29.83, $p = .051$, 95% CI [-59.33, 0.50]; incongruent, $F(1, 181) = 4.09$, $p = .045$, mean difference = -36.58, $p = .049$, 95% CI [-72.23, -0.61] (Figure A.4), but there was no difference between the groups on an RT cost score (i.e. incongruent trial RT – congruent trial RT), $F(1, 181) < 1$. Further, there were no differences between the groups on error rate (ER) for congruent, $F(1, 181) < 1$, or incongruent trial types, $F(1, 181) = 1.21$, $p = .272$, again suggesting differences in RT were not due to a speed accuracy tradeoff.

When cardiorespiratory fitness level was added into the model, the sample size was reduced to N = 153 with allelic group sizes as follows: C/C Homozygotes = 105, T Carriers = 48. The interaction between DBH -1021 C/T x Fitness Level did not significantly explain any variance in RT or ER for either congruent or incongruent trial types (Table 3.7).

Wisconsin card sort test (WCST). The sample size was N = 182 with allelic group sizes as follows: C/C Homozygotes = 131, T Carriers = 51. There were no group differences on performance of the WCST measured by total errors or percent perseverative errors, both $F(1, 176) < 1$. When cardiorespiratory fitness level was added into the model, the sample size was reduced to N = 150 with allelic group sizes as follows: C/C Homozygotes = 107, T Carriers = 43. The interaction significantly explained 3.1% of the variance in total errors, $\Delta R^2 = .031$, $\Delta F(1, 142) = 5.28$, $p = .023$. Simple slopes analysis showed WCST total errors significantly decreased with increasing cardiorespiratory fitness levels for the T Carriers, B = -2.10, β = -0.95, $t(142) = -2.58$, $p = .004$, 95% CI [-3.57, -0.64], but not for C/C Homozygotes, B = -0.13, β = -0.12, $t(142) = -0.24$, $p = .809$, 95% CI [-1.21, 0.84] (Figure 3.4a). The interaction also marginally explained 2.1% of the variance in percent perseverative errors, $\Delta R^2 = .021$, $\Delta F(1, 142) = 3.33$, $p = .070$. However, simple slopes analysis revealed a non-significant relationship between percent perseverative errors and cardiorespiratory fitness level for both T Carriers, B = -0.65, β = -0.78, $t(142) = -2.05$, $p = .109$, 95% CI [-1.53, 0.11], and C/C Homozygotes, B = -0.04, β = -0.01, $t(142) = -0.20$, $p = .851$, 95% CI [-0.47, 0.36]. Further, the bootstrapped regression results showed the slopes

were not significantly different from each other, B = -0.61, β = -0.73, $t(142)$ = -1.82, p = .156, 95% CI [-1.48, 0.19] (Figure 3.4b).

Task switch test. The sample size was N = 156 with allelic group sizes as follows: C/C Homozygotes = 107, T Carriers = 50. There were no significant differences between the allelic groups on response time (RT) for the single condition, $F(1, 151) < 1$, but the T Carriers were significantly faster during the mix condition $F(1, 151) = 6.20$, p = .014, mean difference = -62.63, p = .016, 95% CI [-113.36, -13.37] (Figure A.5a). Furthermore, T Carriers showed a significantly lower global RT cost (i.e. mix condition RT – single condition RT), $F(1, 151) = 5.21$, p = .024, mean difference = -50.71, p = .014, 95% CI [-92.19, -9.81] (Figure A.5b). There were no allelic differences on error rate (ER) for either the single condition, $F(1, 151) < 1$, or the mix condition, $F(1, 151) = 1.85$, p = .176 (Figure A.5c). However, there was a marginal allelic effect on a global error cost score (i.e. mix condition ER – single condition ER), $F(1, 151) = 3.88$, p = .051, with T Carriers showing a lower cost to the mix compared to the single condition in terms of error rate compared to the C/C group, mean difference = -.037, p = .058, 95% CI [-.075, .001] (Figure A.5d). Thus T Carriers were not only faster, but also more accurate overall.

Further analysis of the task switch test mix condition showed T Carriers were marginally faster than C/C Homozygotes on RT for repeat trials, $F(1, 151) = 2.97$, p = .087, mean difference = -42.95, p = .079, 95% CI [-89.77, 6.16], and significantly faster on switch trials, $F(1, 151) = 6.28$, p = .013, mean difference = -82.31, p = .017, 95% CI [-146.96, -15.73] (Figure A.6a). There were no differences on ER for either repeat trials, $F(1, 151) = 2.50$, p = .116, or switch trials, $F(1, 151) = 1.20$, p = .276. Further, there were no group differences on local cost scores (i.e. switch trial – repeat trial) for either RT, $F(1, 151) = 1.77$, p = .184, or ER, $F(1, 151) < 1$. T Carriers were faster and more accurate overall, suggesting they are better at maintaining and switching between two memory sets.

When cardiorespiratory fitness level was added into the model, the sample size was reduced to N = 131 with allelic group sizes as follows: C/C Homozygotes = 88, T Carriers = 43. The interaction between DBH -1021 C/T X Fitness Level did not show a significant effect on RT or ER for any condition or trial type (Table 3.8).

Dual task test. The sample size for the dual task test was $N = 158$ with allelic group sizes as follows: C/C Homozygotes = 108, T Carriers = 50. T Carriers showed significantly faster response times (RT) on both single trials, $F(1, 152) = 4.82$, $p = .030$, mean difference = -62.06, $p = .014$, 95% CI [-113.51, -13.48], and marginally on dual trials, $F(1, 152) = 2.77$, $p = .098$, mean difference = -55.11, $p = .091$, 95% CI [-118.77, 9.57] (Figure A.7). Further, there were no differences between the groups on error rate (ER) for either single or dual trials, both $F(1, 152) < 1$, indicating there was not a speed accuracy tradeoff. There were also no group differences on cost scores (i.e. dual trial – single trial) RT, $F(1, 153) < 1$, or ER, $F(1, 152) = 1.38$, $p = .242$.

When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 151$ with allelic group sizes as follows: C/C Homozygotes = 103, T Carriers = 48. The interaction again did not show a significant effect on RT or ER for any trial type (Table 3.9).

DBH Combined SNPs

To further study the effects of the DBH gene on cognition in healthy older adults, I conducted an analysis using a combination of the two SNPs. I created a DBH Low DA group (i.e. high DBH translating to low DA:NE) which included those that were G/G Homozygotes on DBH 444 G/A and C/C Homozygotes on DBH -1021 C/T. I compared this group to a DBH High DA group (i.e. low DBH translating to high DA:NE) which included those that were A/A Homozygotes on the DBH 444 G/A SNP and carried the T allele on DBH -1021 C/T.

There were no significant differences between the DBH Low DA and DBH High DA groups on age, education, BMI or VO_2; however, the groups did significantly differ on mMMSE (Table 3.10). Chi-square analysis revealed a marginal difference on the distribution of males and females in the groups. For the DBH Low DA group, there were more females and fewer males than expected, while the opposite was true for the DBH High DA group. However, this difference should not affect the results since sex was already planned to be used as control variable. Due to the difference found in mMMSE, it was also added as a control variable for the following analyses. Means and standard deviations for the primary measures on the behavioral tests are reported in Table 3.11.

Verbal crystallized intelligence. Since verbal crystallized intelligence remains relatively stable in older adult populations (Hedden & Gabrieli, 2004), I do not expect to find a significant allelic or interaction effect on performance for any of following measures of verbal crystallized intelligence.

KBIT test. The sample size for the KBIT test was $N = 49$, with group sizes as follows: DBH Low DA = 32 and DBH High DA = 17. There were no group differences on the age-scaled scores for the K-BIT test, $F(1, 43) = 1.44$, $p = .237$. When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 41$ with group sizes as follows: DBH Low DA = 28 and DBH High DA = 13. The interaction between DBH x Fitness Level did not significantly explain any variance in age-scaled scores, $\Delta R^2 = .000$, $\Delta F(1, 33) < 1$.

Memory tests. Performance on working and spatial memory tests decrease in older adults, in contrast to general sparing in tests of short term memory (Hedden & Gabrieli, 2004; Park et al., 2002). I predict that there will be no effect of DBH gene status on the test of short-term memory (i.e. forward span), but that the DBH High DA group will out perform the DBH Low DA group on the tests of working (i.e. backward span) and spatial memory due to more availability of prefrontal dopamine. I further hypothesize an interactive effect for both the working and spatial memory test such that there will be a larger effect of cardiorespiratory fitness level in the DBH Low DA group, with no effect on the test of short term memory.

Digit span test. The sample size for the digit span test was $N = 49$, with group sizes as follows: DBH Low DA = 32, DBH High DA = 17. There were no differences between groups on the length of span for either the forward span, $F(1, 43) < 1$, or the backward span, $F(1, 43) = 1.63$, $p = .208$.

When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 41$ with group sizes as follows: DBH Low DA = 28, DBH High DA = 13. The interaction between DBH x Fitness Level did not significantly explain any variance in the forward span, $\Delta R^2 = .000$, $\Delta F(1, 33) < 1$, or backward span, $\Delta R^2 = .052$, $\Delta F(1, 33) = 2.31$, $p = .097$.

Spatial memory test. The sample size was $N = 49$, with group sizes as follows: DBH Low DA = 28, DBH High DA = 16. The DBH High DA group showed a trend towards faster response times (RT) compared to the DBH Low DA group across all memory loads: memory load 1 RT, $F(1, 38) = 2.55$, $p = .118$, mean difference = -85.66, $p = .094$, 95% CI [-184.78, 9.90]; memory load 2 RT, $F(1, 38) = 3.36$, $p = .074$, mean difference = -91.35, $p = .080$, 95% CI [-190.59, 7.11]; memory load 3 RT, $F(1, 38) = 3.66$, $p = .063$, mean difference = -104.08, $p = .063$, 95% CI [-214.27, 6.11]. Further, there were no significant differences on error rate (ER) for any memory load: memory load 1, $F(1, 38) < 1$; memory load 2, $F(1, 38) = 1.41$, $p = .243$; memory load 3, $F(1, 38) < 1$, suggesting potential differences in RT were not due to speed accuracy tradeoffs.

When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 36$ with group sizes as follows: DBH Low DA = 24, DBH High DA = 12. The interaction between DBH x Fitness Level had no significant effects on RT or ER for any memory load (Table 3.12).

Executive control function tests. Aging is thought to have the greatest negative effect on performance of tests of executive control (Hedden & Gabrieli, 2004; Park & Gutchess, 2002). Both the G allele for DBH 444 G/A and the C allele for the DBH -1021 C/T are thought to result in less DBH activity (Hedden & Gabrieli, 2004; Park & Gutchess, 2002) implying a lower DA:NE ratio which may lead to worse performance on some measures of executive control (Greene et al., 2009). In contrast, higher levels of cardiorespiratory fitness are associated with better performance on measures of executive control (Colcombe & Kramer, 2003; Kramer & Erickson, 2007). Therefore, I expect the DBH High DA group will outperform the DBH Low DA group, but that there will be a positive effect of cardiorespiratory fitness level within the DBH Low DA group.

Flanker test. For the Flanker test, the sample size was $N = 47$ with group sizes as follows: DBH Low DA = 32, DBH High DA = 15. As predicted, the DBH High DA group was significantly faster than the DBH Low DA group on both trial types: congruent, $F(1, 41) = 4.96$, $p = .031$, mean difference = -77.53, $p = .058$, 95% CI [-158.23, -3.81]; incongruent, $F(1, 41) = 5.11$, $p = .029$, mean difference = -89.48, $p = .045$, 95% CI [-179.08, -13.43] (Figure A.8). Further, there were no differences between the groups on error rate (ER) for

congruent trials, $F(1, 41) = 1.04$, $p = .314$, or incongruent trial types, $F(1, 41) = 1.84$, $p = .182$, suggesting differences in RT were not due to a speed accuracy tradeoff. However, there was no difference on a RT cost score (i.e. incongruent RT – congruent RT), $F(1, 41) < 1$, suggesting that the DBH gene may influence overall performance of the task and not specifically the more demanding incongruent trial types.

When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 40$ with group sizes as follows: DBH Low DA = 28, DBH High DA = 12. The interaction between DBH x Fitness Level did not significantly explain any variance in RT or ER for either congruent or incongruent trial types (Table 3.13).

Task switch test. For the task switch test, the sample size was $N = 42$ with group sizes as follows: DBH Low DA = 25, DBH High DA = 17. ANCOVA showed no group differences on response time (RT) or error rate (ER) for either the single or the mix condition (Table A.2). However, the DBH High DA group showed a marginally lower global ER cost (i.e. mix condition ER – single condition ER), $F(1, 36) = 3.17$, $p = .083$, mean difference = -.074, $p = .094$, 95% CI [-.161, .005] (Figure A.9).

Further analysis of the task switch test mix condition showed no significant group differences on RT or ER for either repeat or switch trials (Table A.2). There was no difference on a local cost score (i.e. switch trial – repeat trial) for either RT, $F(1, 36) = 1.24$, $p = .272$, or ER, both $F(1, 36) < 1$.

When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 41$ with group sizes as follows: DBH Low DA = 28, DBH High DA = 13. The interaction between DBH X Fitness Level did not show a significant effect on RT or ER for either condition (Table 3.14). Further analysis of the mix condition showed no differences between the groups on RT or ER for either trial type (Table 3.14). However, the interaction did significantly explain 10.2% of the variance in local ER cost (i.e. switch trial ER – repeat trial ER), $\Delta R^2 = .102$, $\Delta F(1, 27) = 4.83$, $p = .037$. Simple slopes analysis revealed local ER cost decreased with increasing VO_2 for the DBH low DA group, $B = -0.012$, $\beta = -1.84$, $t(27) = -3.57$, $p = .001$, 95% CI [-.019, -.005], but not the DBH High DA group, $B = -0.003$, $\beta = -0.47$, $t(27) = -0.80$, $p = .430$, 95% CI [-.011, .005] (Figure 3.18b).

Catechol-O-Methyltransferase

The Catechol-O-Methyltransferase (COMT) gene encodes for a methylation enzyme that metabolizes catecholamines including dopamine (Axelrod & Tomchick, 1958). Although the enzyme has a wide distribution throughout the body and brain, it plays a key role in the prefrontal cortex where there is a low level of dopamine transporters (DAT; Chen et al., 2004). Therefore, prefrontal synaptic dopamine (DA) is mostly inactivated by diffusion, receptor internalization, and COMT action. This is evidenced by studies in COMT-knockout mice that show increased DA levels in the PFC, in contrast to normal DA levels in the striatum, a brain area where DATs are abundant (Gogos et al., 1998; Karoum, Chrapusta, & Egan, 1994). Since COMT seems to control the basal DA neurotransmission levels within the PFC, it is a great candidate for possible individual differences in human behavior. To investigate this possibility, two common SNPs have been studied in the COMT gene, COMT Val158Met (rs4680), which affects enzymatic activity, and COMT C/G (rs4818), which affects enzyme availability.

COMT Val158Met is a functional SNP that causes a methionine (Met) to replace a valine (Val) in the amino acid sequence at codon 158. This change to Met produces a less thermostable enzyme, such that the Met/Met variant shows 40% less enzymatic activity compared to the Val/Val, resulting in higher PFC DA levels (Chen et al., 2004; Lotta et al., 1995; Savitz et al., 2006). Through positron emission tomography, Slifstein et al. (2008) showed that young adult Val homozygotes had higher levels of D1 receptor binding in cortical and limbic regions compared to Met allele carriers, indicative of lower DA tone. This differential effect was not found in striatal regions as would be predicted due to the differential distribution of the DAT between cortical and striatal regions.

Another SNP, COMT C/G, affects mRNA secondary structure, consequently affecting protein synthesis, but not the functioning of the enzyme itself (Nackley et al., 2006). Carriers of the C allele are believed to have lower COMT activity due to less efficient protein synthesis (Diatchenko et al., 2005; Nackley et al., 2006). However, to date, there have not been any studies investigating the differential effects of the COMT C/G SNP on levels of DA in the human brain.

COMT Predictions

In relation to behavior, due to the higher activity COMT, Val allele carriers for the COMT Val158Met are hypothesized to perform worse on PFC-mediated executive control tasks (Chen et al., 2004; Egan et al., 2001; Goldberg et al., 2003; Malhotra et al., 2002; Mattay & Goldberg, 2004). Many studies have shown this relationship between the COMT Val158Met genotype and executive performance such that Met homozygotes do tend to outperform Val homozygotes on tasks requiring executive processing specifically and not tasks in other domains (Egan et al., 2001; Goldberg et al., 2003; Malhotra et al., 2002; Nagel et al., 2008; Savitz et al., 2006). For example, Egan et al. (2001) showed Val homozygotes committed significantly more perseverative errors compared to Met homozygotes on the Wisconsin Card Sorting Task (WCST), which showed an inability to flexibly adapt to a new rule. This effect was similar in normal subjects and schizophrenic patients, indicating the effect of variation was not related to the disease, but rather a generalizable human characteristic. Further, Goldberg et al. (2003) showed that Val homozygotes, compared to Met homozygotes, were significantly less accurate on the 1-back and 2-back conditions of an *N*-back task, revealing a deficit in working memory. Mattay et al. (2004) also showed that Val homozygotes made more perseverative errors on the WCST, but were matched on performance at all levels (1, 2, and 3-back) of an *N*-back task with Met homozygotes. Interestingly, administration of amphetamine (AMP; which increases PFC dopamine levels) to Val homozygotes improved response time, but not accuracy on the 3-back version of the *N*-back task and decreased perseverative errors on the WCST. In contrast, AMP administration to Met homozygotes diminished performance on both the 3-back task and WCST. This data is consistent with evidence that DA impacts PFC function in an inverted U-shaped curve, and shows that the COMT Val158Met genotype can modify the relationship between dopamine availability in the PFC and executive functioning.

The majority of studies have used young to middle-aged participants, but the question then arises as to whether the same results would be found in older adults due to aging-related declines in the DA system. Addressing this question, Nagel et al. (2008) showed that human aging magnifies the effect of the COMT Val158Met genotype on executive functioning such that only older adult Val homozygotes showed a higher number

of perseverative errors on the WCST. An association between COMT Val158Met genotype and number of perseverative errors was not found in their younger adult population.

There are fewer studies exploring the effects of the COMT C/G polymorphism on cognition; however, Roussos et al. (Roussos, Giakoumaki, & Bitsios, 2009; Roussos, Giakoumaki, Pavlakis, & Bitsios, 2008) has found that the G/G genotype performs worse on tasks of executive control. In 2008, they found that young healthy males with the G/G genotype performed worse on the Stockings of Cambridge test (a modified, computerized version of the Tower of London), but better on the Iowa Gambling Test (a test requiring emotionally informed decision-making). In 2009, again using healthy young males, they found that the G/G genotype had a slower response time in an *N*-back test compared with the C/C genotype, and that Tolcapone (a COMT inhibitor) improved response times only for the G/G group.

Although results have been equivocal, it is apparent that the COMT genotype is playing some role in PFC mediated cognitive functioning and this relationship may increase as a function of age. Therefore, I predict that in the current study there will be an interaction between Fitness Level and COMT genotypes such that the Val carriers and G carriers will show a positive relationship between cardiorespiratory fitness level and behavior, but the other genotypes will not.

COMT Results

COMT Val158Met (rs4680)

There were no significant differences between the Val carriers and Met/Met Homozygotes on age, education, mMMSE, Body Mass Index (BMI) or VO_2 (Table 3.14). Chi-square analysis showed no differences in the distribution of males and females between the groups. Means and standard deviations for the primary measures on the behavioral tests are reported in Table 3.15

Verbal crystallized intelligence. Verbal crystallized intelligence is believed to remain relatively stable in older adults (Hedden & Gabrieli, 2004; Park & Gutchess, 2002). I

hypothesize that there will be no significant differences between the genotypic groups on performance of the KBIT test, nor a significant interaction effect with fitness level.

KBIT Test. The sample size for the KBIT test was N = 193 with the genotypic group sizes as follows: Val/Val = 31, Val/Met = 105, Met/Met = 57. Analysis of covariance (ANCOVA) showed no differences between the genotypes, $F(2, 187) < 1$, suggesting COMT Val158Met does not affect verbal crystallized intelligence. When cardiorespiratory fitness level was added to the model, the sample size for the KBIT test was reduced to $N = 158$ with genotypic group sizes as follows: Val/Val = 27, Val/Met = 85, Met/Met = 46. Hierarchical regression showed the interaction between COMT Val158Met X Fitness Level did not significantly explain any variance in age-scaled scores, $\Delta R^2 = .009$, $\Delta F(2, 149) < 1$.

Memory tests. Performance on working and spatial memory tests decrease in older adults, in contrast to general sparing in tests of short-term memory (Hedden & Gabrieli, 2004; Park & Gutchess, 2002). In terms of the COMT Val158Met SNP, Val carriers are believed to have lower prefrontal dopamine levels and perform worse on working and spatial memory measures compared to Met/Met homozygotes. Therefore, I predict there will be no genotypic effect on the test of short-term memory, but the Met/Met homozygotes will outperform any Val carriers. Furthermore, higher cardiorespiratory fitness levels are associated with better performance on measures of working and spatial memory. Therefore, I expect there will be no Genotype x Fitness Level interaction effect on the test of short-term memory, but the Val/Val homozygotes will show better performance on the measures of short term and spatial memory with increasing cardiorespiratory fitness levels.

Digit span test. The sample size for the digit span test was N = 195, with the genotypic group sizes as follows: Val/Val = 31, Val/Met = 107, Met/Met = 57. There were no significant differences between the genotypic groups on forward span, $F(2, 189) = 1.05$, $p = .352$ (Figure A.10). However, there was a marginal genotypic effect on the backward span, $F(2, 189) = 2.45$, $p = .089$ (Figure A.10). Bootstrapped pairwise comparisons revealed the Val/Val Homozygotes repeated more digits compared to both the Val/Met heterozygotes, mean difference = 0.53, $p = .048$, 95% CI [0.02, 1.01] and the Met/Met homozygotes, mean difference = 0.60, $p = .040$, 95% CI [0.04, 1.19]. There was not a

significant difference between the Val/Met heterozygotes and the Met/Met homozygotes, mean difference = .067, p = .787, 95% CI [-0.32, 0.47]. These results were inconsistent with the prediction.

When cardiorespiratory fitness level was added into the model, the sample size was reduced to N = 160, with genotypic group sizes as follows: Val/Val = 27, Val/Met = 87, Met/Met = 46. As expected, the interaction did not explain a significant amount of variance for the forward span, ΔR^2 = .016, $\Delta F(2, 151) = 1.22$, p = .271 (Figure 3.6a), but did significantly explain 5.4% of the variance for the backward span, ΔR^2 = .054, $\Delta F(2, 151) = 4.99$, p = .008 (Figure 3.6b). Simple slopes analysis revealed a significant relationship between the number of digits and cardiorespiratory fitness level only for the Val/Val group, B = 0.24, β = .272, $t(151) = 3.20$, p = .000, 95% CI [0.09, 0.36], while the other two genotypic groups did not show a significant relationship, Val/Met, B = -0.00, β = -.010, $t(151) = -0.12$, p = .919, 95% CI [-0.07, 0.08], and Met/Met, B = 0.00, β = .000, $t(151) = -0.01$, p = .995, 95% CI [-0.08, 0.09]. These results followed prediction that increasing fitness level would specifically influence the Val/Val Homozygotes.

Spatial memory test. The sample size for the spatial memory test was N = 175, with the genotypic group sizes as follows: Val/Val = 28, Val/Met = 96, Met/Met = 51. Analysis of covariance (ANCOVA) showed COMT Val158Met had a significant effect on response time (RT) for all memory loads: memory load 1, $F(2, 169) = 4.81$, p = .009; memory load 2, $F(2, 169) = 4.13$, p = .018; memory load 3, $F(2, 169) = 6.41$, p = .002 (Figure A.11). Bootstrapped pairwise comparisons revealed the Met/Met homozygotes were significantly slower than both the Val/Val Homozygotes and Val/Met heterozygotes (Table A.3). There were no group differences on error rate (ER) for any memory load, suggesting there was no speed accuracy trade off: memory load 1, $F(2, 169) < 1$; memory load 2, $F(2, 169) = 1.04$, p = .355; memory load 3, $F(2, 169) < 1$.

When cardiorespiratory fitness level was added into the model, the sample size for the Spatial Memory test was reduced to N = 143 with genotypic group sizes as follows: Val/Val = 25, Val/Met = 77, Met/Met = 41. Although, a genotypic effect was seen, the interaction between COMT Val158Met x Fitness Level did not have a significant effect on either RT or ER for any memory load of the spatial memory test (Table 3.17).

Executive control function tests. Older adults have been shown to have decreasing performance on tests of executive control function , but higher levels of fitness tend to be associated with better executive control (Bherer, Erickson, & Liu-Ambrose, 2013; Colcombe & Kramer, 2003; Prakash et al., 2015). The Val allele is biologically associated with lower levels of prefrontal dopamine and behaviorally with worse performance on tests of executive control (Goldberg & Weinberger, 2004). Therefore, I expect a significant effect of COMT Val158Met Genotype such that Met/Met homozygotes will outperform Val carriers. Further, I expect cardiorespiratory fitness level to have a greater affect on performance in Val carriers compared to Met/Met homozygotes.

Wisconsin card sorting test (WCST). The sample size for the Wisconsin Card Sorting Test was N = 181 with genotypic group sizes as follows: Val/Val = 28, Val/Met = 98, Met/Met = 55. Against prediction, ANCOVA showed COMT Val158Met did not have an effect on either total errors, $F(2, 175) < 1$, or percent perseverative errors, $F(2, 175) = 1.47$, $p = .234$.

When cardiorespiratory fitness level was added to the model, the sample size for the WCST was reduced to $N = 149$, with genotypic group sizes as follows: Val/Val = 24, Val/Met = 81, Met/Met = 44. The interaction did not significantly explain any variance for the two measures of the WCST: total errors, $\Delta R^2 = .019$, $\Delta F(2, 140) = 1.53$, $p = .221$; percent perseverative errors, $\Delta R^2 = .029$, $\Delta F(2, 140) = 2.26$, $p = .108$.

Flanker test. The sample size for the Flanker test was $N = 186$, with genotypic group sizes as follows: Val/Val = 30, Val/Met = 101, Met/Met = 55. There was a marginal effect of the COMT Val158Met genotype on response time for congruent trials, $F(2, 180) = 2.55$, $p = .081$, and a significant effect for incongruent trials, $F(2, 180) = 3.47$, $p = .033$ (Figure A.12a). Unexpectedly, bootstrapped pairwise comparisons revealed that Met/Met homozygotes had the slowest response times, and the Val/Val Homozygotes were fastest, for both congruent and incongruent trials (Table A.4). There was not a significant difference for ER for congruent trials, $F(2, 180) = 1.45$, $p = .238$, however, there was a significant group difference on error rate (ER) for incongruent trials, $F(2, 180) = 4.63$, $p = .011$ (Figure A.12c), with the Val/Val group showing the highest ER compared to the other groups (Table A.4). There was also a group difference on an ER cost (i.e. Incongruent ER – Congruent ER), $F(2, 180) = 3.86$, $p = .023$ (Figure A.12d). This could be an indication of the

Val/Val homozygotes trading speed for accuracy. There was no significant genotypic effect on an RT cost score (i.e. incongruent RT – congruent RT), $F(2, 180) = 1.15$, $p = .320$ (Figure A.12b).

When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 152$ with genotypic group sizes as follows: Val/Val = 27, Val/Met = 81, Met/Met = 44. The interaction between COMT Val158Met x Fitness Level marginally explained 3.5% of the variance in RT for congruent trials, $\Delta R^2 = .035$, $\Delta F(2, 143) = 2.90$, $p = .058$. The results suggest there was a difference in the direction of the relationship between response and cardiorespiratory fitness level between the groups: Val/Val vs Val/Met, $B = 10.24$, $\beta = .403$, $t(143) = 2.06$, $p = .060$, 95% CI [0.43, 22.87]; Val/Val vs Met/Met, $B = 12.61$, $\beta = .383$, $t(143) = 2.40$, $p = .031$, 95% CI [1.74, 25.63] (Figure 3.7). However, simple slopes analysis showed there was a non-significant relationship between response time and fitness level on congruent trials for each of the genotypic groups (Table 3.19). The interaction did not explain variance in RT for incongruent trials or ER for either congruent or incongruent trial types (Table 3.18). xxx

Dual task test. The sample size for the dual task test was $N = 157$, with the genotypic group sizes as follows: Val/Val = 27, Val/Met = 83, Met/Met = 47. ANCOVA showed significant differences between the genotypic groups on response time (RT) for both single, $F(2, 157) = 4.07$, $p = .019$, and dual trial types, $F(2, 151) = 5.50$, $p = .005$, with Val/Val homozygotes being the fastest (Figure A.13a; Table A.5). There were also differences between the groups on error rate (ER) for both single, $F(2, 157) = 2.99$, $p = .054$, and dual trial types, $F(2, 157) = 4.22$, $p = .017$ (Figure A.13c; Table A.5). Val/Val homozygotes showed lower error rates, suggesting there was not a speed accuracy tradeoff. There were no group differences on an RT cost score (i.e. dual RT – single RT), $F(2, 151) = 2.16$, $p = .118$ (Figure A.13b), but there was a marginal difference on an ER cost score (i.e. dual ER – single ER), $F(2, 151) = 2.44$, $p = .091$. Bootstrapped pairwise comparisons showed the Val/Val group had the lowest ER cost (Figure A.13d; Table A.5).

When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 150$, with genotypic group sizes as follows: Val/Val = 26, Val/Met = 81, Met/Met = 43. For response time, the interaction between COMT Val158Met x Fitness Level did not significantly explain any variance for either single trials, $\Delta R^2 = .002$, $\Delta F(2, 141) < 1$,

or dual trials, $\Delta R^2 = .010$, $\Delta F(2, 141) < 1$. However, the interaction did marginally explain 3.1% of variance in a RT cost score (i.e. dual RT – single RT), $\Delta R^2 = .031$, $\Delta F(2, 141) = 2.55$, $p = .082$ (Figure 3.8). Simple slopes analysis revealed an odd result suggesting cost significantly increased with increasing VO_2 for the Val/Met heterozygotes, $B = 7.99$, $\beta = .222$, $t(141) = 2.37$, $p = .025$, 95% CI [0.70, 14.75], but not for the Val/Val group, $B = 0.86$, $\beta = .012$, $t(141) = 0.13$, $p = .873$, 95% CI [-10.14, 11.17], or the Met/Met group, $B = -2.20$, $\beta = -.048$, $t(141) = -0.54$, $p = .572$, 95% CI [-9.40, 5.78]. Further, the slope for the Val/Met group was significantly different than the Met/Met group, $B = -10.19$, $\beta = -.223$, $t(141) = -2.20$, $p = .031$, 95% CI [-19.27, -0.75], but not different than the Val/Val group, $B = -7.13$, $\beta = -.10$, $t(141) = -1.01$, $p = .217$, 95% CI [-20.43, 4.81].

COMT C/G (rs4818)

There were no significant differences between the G Carriers and C/C Homozygotes on age, education, mMMSE, or VO_2 (Table 3.20). The C/C Homozygotes did have a significantly lower BMI, t(82) = 2.71, p = .008. Chi-square analysis showed no differences in the distribution of males and females between the groups. Means and standard deviations for the primary measures on the behavioral tests are reported in Table 3.21.

Verbal crystallized intelligence. Verbal crystallized intelligence is believed to remain relatively stable among older adults (Hedden & Gabrieli, 2004; D.C Park & Gutchess, 2000). I hypothesize there will be no differences on performance of any of the following measures between the genotypic groups for the COMT C/G SNP.

***KBIT* test.** The sample size for the KBIT test was $N = 193$, with genotypic group sizes as follows: G Carriers = 113, C/C Homozygotes = 80. Following prediction, analysis of covariance (ANCOVA) showed no significant differences between the genotypic groups on age-scaled scores, $F(1, 188) < 1$.

When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 158$, with allelic group sizes as follows: G Carriers = 92, C/C Homozygotes = 66. Again following prediction, hierarchical regression showed the interaction between COMT C/G x Fitness Level did not significantly explain any variance in the age-scaled scores, $\Delta R^2 = .000$, $\Delta F(1, 151) < 1$.

Memory tests. Performance on working and spatial memory tests decreases in older adults, in contrast to general sparing in tests of short term memory (Hedden & Gabrieli, 2004; D.C Park & Gutchess, 2000). In terms of the COMT C/G SNP, G carriers have been shown to have lower prefrontal dopamine levels and perform worse on working and spatial memory measures compared to C/C Homozygotes. Therefore, I predict there will be no genotypic effect on the test of short-term memory, but the C/C Homozygotes will perform better than G Carriers on the tests of working and spatial memory.

Digit span test. The sample size was $N = 195$ with allelic group sizes as follows: G Carriers = 115, C/C Homozygotes = 80. Analysis of covariance (ANCOVA) showed there were no differences between the groups on forward span, $F(1, 190) < 1$, but for backward span, G Carriers had marginally longer spans, $F(1, 190) = 3.36$, $p = .068$, mean difference = 0.34, $p = .068$, 95% CI [-0.03, 0.71] (Figure A.14). When cardiorespiratory fitness level was added into the model, the COMT C/G x Fitness Level interaction did not explain variance in the forward span, $\Delta R^2 = .002$, $\Delta F(1, 153) = .321$, $p = .572$, or backward span, $\Delta R^2 = .007$, $\Delta F(1, 153) = .1.27$, $p = .262$.

Spatial memory test. The sample size for the spatial memory test was $N = 175$ with allelic group sizes as follows: G Carriers = 105, C/C Homozygotes = 70. ANCOVA showed G Carriers had significantly faster response times (RT) for all memory loads: memory load 1, $F(1, 170) = 5.00$, $p = .027$, mean difference = -57.87, $p = .029$, 95% CI [-109.13, -6.10]; memory load 2, $F(1, 170) = 4.21$, $p = .042$, mean difference = -52.28, $p = .045$, 95% CI [-103.64, -2.05]; memory load 3, $F(1, 170) = 7.71$, $p = .006$, mean difference = -76.46, $p = .010$, 95% CI [-134.05, -20.85] (Figure A.15). There were no differences on error rate for any memory load, suggesting there was no speed accuracy tradeoff, all $F(1, 170) < 1$ (Table 3.22).

When cardiorespiratory fitness level was added into the model, the sample size for the spatial memory test was reduced to $N = 143$, with genotypic samples sizes as follows: G Carriers = 85, C/C Homozygotes = 58. Hierarchical regression showed the interaction did not significantly explain any variance for either RT or ER for any memory load (Table 3.22).

Executive control function tests. Older adults have been shown to have decreasing performance on tests of executive control function (Hedden & Gabrieli, 2004; D.C Park &

Gutchess, 2000). Carriers of the G allele are believed to have the lower prefrontal dopamine levels and perform worse on measures of executive control (Roussos et al., 2009, 2008). Therefore, I expect a significant effect of COMT C/G genotype such that C/C Homozygotes will outperform G Carriers.

Flanker test. The sample size for the Flanker test was N = 186, with genotypic group sizes as follows: G Carriers = 110, C/C Homozygotes = 76. ANCOVA showed the G Carriers were significantly faster than C/C Homozygotes on congruent trials, $F(1, 181) = 5.22$, p = .023, mean difference = -29.78, p = .024, 95% CI [-56.53, -3.90], and marginally faster on incongruent trials, $F(1, 181) = 3.73$, p = .055, mean difference = -31.14, p = .063, 95% CI [-64.03, 1.35] (Figure A.16). However, there was no difference seen on an RT cost score (i.e. incongruent RT – congruent RT), $F(1, 181) < 1$. There was also no difference seen on error rate for either congruent or incongruent trials, both $F(1, 181) < 1$. Against prediction, these results suggest G Carriers are faster overall.

When cardiorespiratory fitness level was added into the model, the sample size for the Flanker test was reduced to N = 152, with genotypic group sizes as follows: G Carriers = 90, C/C Homozygotes = 62. Hierarchical regression showed the interaction did not significantly explain any variance in RT or ER for either trial type (Table 3.23).

Wisconsin card sort. The sample size for the WCST was N = 181, with allelic group sizes as follows: G Carriers = 105, C/C Homozygotes = 76. ANCOVA showed no differences between the groups on either total errors or percent perseverative errors, both $F(1, 176) < 1$.

When cardiorespiratory fitness level was added into the model, the sample size for the WCST was reduced to N = 149, with allelic group sizes as follows: G Carriers = 86, C/C Homozygotes = 63. Hierarchical regression showed the interaction between COMT C/G x Fitness Level did not significantly explain any variance in performance total errors ΔR^2 = .014, $\Delta F(1, 142) = 2.22$, p = .139. However, the interaction did significantly explain 3% of the variance in percent perseverative errors, ΔR^2 = .030, $\Delta F(1, 142) = 4.68$, p = .032 (Figure 3.9). Simple slopes analysis showed that percent perseverative errors marginally decreased with increasing VO_2 only in the C/C group, B = -0.54, β = -.203, $t(142) = -2.22$, p =

.052, 95% CI [-1.11, -0.29], and not the G Carriers, B = 0.10, β = .041, $t(142)$ = .430, p = .671, 95% CI [-0.37, -0.59].

Dual task test. The sample size for the dual task test was N = 157, with allelic group sizes as follows: G Carriers = 91, C/C Homozygotes = 66. ANCOVA showed G Carriers were significantly faster during dual trials, $F(1, 152)$ = 4.86, p = .029, mean difference = -66.91, p = .029, 95% CI [-126.78, -8.42], but there were no difference single trials, $F(1, 152) < 1$ (Figure A.17a). Further, G Carriers also showed a significantly lower RT cost (i.e. dual RT – single RT), $F(1, 152)$ = 5.71, p = .018, mean difference = -46.95, p = .018, 95% CI [-84.77, -9.17] (Figure A.17b). There were no differences seen in error rate for either single or dual trials, both $F(1, 152) < 1$), suggesting there was no speed accuracy tradeoff.

When cardiorespiratory fitness level was added into the model, the sample size for the dual task test was reduced to N = 150, with genotypic group sizes as follows: G Carriers = 89, C/C Homozygotes = 61. Hierarchical regression showed the interaction between COMT C/G x Fitness Level did not significantly explain any variance in response time in either the single trials, ΔR^2 = .003, $\Delta F(1, 143) < 1$, or dual trials, ΔR^2 = .004, $\Delta F(1, 143) < 1$. However, the interaction did significantly explain 2.9% of the variance in a RT cost score (i.e. dual RT – single RT), ΔR^2 = .029, $\Delta F(1, 143)$ = 4.74, p = .031 (Figure 3.10). Simple slopes analysis showed RT cost increased with increasing VO_2 for the G Carriers, B = 8.30, β = .230, $t(143)$ = 2.46, p = .011, 95% CI [1.76, 14.59], but there was no relationship for the C/C Homozygotes, B = -0.79, β = -0.02, $t(143)$ = -0.23, p = .835, 95% CI [-8.54, 6.31]. There was no effect on error rate (ER) for either trial type: single, ΔR^2 = .001, $\Delta F(1, 143) < 1$; dual, ΔR^2 = .000, $\Delta F(1, 143) < 1$.

COMT Combined SNPs

To further study the effects of the COMT gene on cognition in healthy older adults, I conducted an analysis using a combination of the two SNPs. I created a high activity COMT (i.e. low prefrontal DA) group which included those that were Val/Val homozygotes on COMT Val158Met and the G allele on COMT C/G. I compared this group to a low activity COMT (i.e. high prefrontal DA) group which included those that were Met/Met homozygotes on the COMT Val158Met and C/C Homozygotes on COMT C/G.

There were no significant differences between the COMT Low DA and COMT High DA groups on age, education, mMMSE, BMI or VO_2 (Table 3.24). Chi-square analysis showed no sex differences between the groups. Means and standard deviations for the primary measures on the behavioral tests are reported in in Table 3.25.

Verbal crystallized intelligence. Verbal crystallized intelligence is believed to remain relatively stable among older adults (Hedden & Gabrieli, 2004; D.C Park & Gutchess, 2000). I hypothesize there will be no group differences on performance of the following test of verbal crystallized intelligence.

***KBIT* test.** The sample size for the KBIT test was $N = 81$, with group sizes as follows: COMT Low DA = 28, COMT High DA = 53. Following prediction, analysis of covariance (ANCOVA) showed there were no significant differences between the genotypic groups on age-scaled scores, $F(1, 81) < 1$.

When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 67$, with group sizes as follows: COMT Low DA = 24, COMT High DA = 43. Again following prediction, hierarchical regression showed the interaction between COMT x Fitness Level did not significantly explain any variance in the age-scaled scores, $\Delta R^2 = .010$, $\Delta F(1, 60) < 1$.

Memory tests. Performance on working and spatial memory tests decrease in older adults, in contrast to general sparing in tests of short term memory (Hedden & Gabrieli, 2004; D.C Park & Gutchess, 2000). In terms of the COMT gene, I expect group differences on the working and spatial memory measures, but not the test of short term memory and expect the low DA group will specifically benefit from increasing fitness level.

Digit span test. The sample size for the digit span test is $N = 81$, with group sizes as follows: COMT Low DA = 28, COMT High DA = 53. As predicted, there were no differences between the groups on the test of short term memory, forward span, $F(1, 76) < 1$, but against prediction, the Low DA group showed significantly better performance on the backward span test of working memory, $F(1, 76) = 5.63$, $p = .020$, mean difference = 0.72, $p = .018$, 95% CI [0.15, 1.31] (Figure A.18).

When cardiorespiratory fitness level was added into the model, the sample size for the spatial memory test was reduced to $N = 67$, with group sizes as follows: COMT Low DA = 24, COMT High DA = 43. Regression showed the interaction between COMT x Fitness Level did not significantly explain variance in forward span, $\Delta R^2 = .013$, $\Delta F(1, 60) = 1.06$, $p = .308$, but did explain 6.6% of the variance in backward span, $\Delta R^2 = .066$, $\Delta F(1, 60) = 5.31$, $p = .025$ (Figure 3.11). Simple slopes analysis showed the number of digits repeated significantly increased with increasing cardiorespiratory fitness level for the Low DA group, $B = 0.18$, $\beta = 1.30$, $t(60) = 2.16$, $p = .042$, 95% CI [-0.02, 0.33], but not for the High DA group, $B = -0.28$, $\beta = -0.22$, $t(60) = -0.59$, $p = .597$, 95% CI [-0.13, 0.08].

Spatial memory test. The sample size for the spatial memory test was $N = 76$, with group sizes as follows: COMT Low DA = 27, COMT High DA = 49. ANCOVA showed the Low DA group had significantly faster response times (RT) for all memory loads: memory load 1, $F(1, 71) = 8.53$, $p = .005$, mean difference = -119.64, $p = .005$, 95% CI [-198.38, -43.13]; memory load 2, $F(1, 71) = 6.17$, $p = .015$, mean difference = -106.56, $p = .013$, 95% CI [-191.22, -26.50]; memory load 3, $F(1, 71) = 8.99$, $p = .004$, mean difference = -139.73, $p = .002$, 95% CI [-223.21, -54.82] (Figure A.19). There were no differences on error rate for any memory load suggesting there was no speed accuracy tradeoff: memory load 1, $F(1, 71) < 1$; memory load 2, $F(1, 71) = 1.24$, $p = .268$; memory load 3, $F(1, 71) < 1$.

When cardiorespiratory fitness level was added into the model, the sample size for the spatial memory test was reduced to $N = 64$, with group sizes as follows: COMT Low DA = 24, COMT High DA = 40. Hierarchical regression showed the interaction did not significantly explain any variance for either RT or ER for any memory load (Table 3.24).

Executive control function tests. Older adults have been shown to have decreasing performance on tests of executive control function (Hedden & Gabrieli, 2004; D.C Park & Gutchess, 2000). Lower prefrontal DA is expected to lead to worse performance on measures of executive control (Roussos et al., 2009, 2008). Therefore, I expect a significant effect of the COMT combined groups such that the COMT High DA group will perform better on the following tasks. Further, I expect cardiorespiratory fitness level will have a greater affect on performance for COMT Low DA group.

Wisconsin card sorting test (WCST). The sample size for the WCST was $N = 77$, with group sizes as follows: COMT Low DA = 26, COMT High DA = 51. ANCOVA showed no differences between the allelic groups on either total errors, $F(1, 72) < 1$ or percent perseverative errors, $F(1, 72) = 1.03$, $p = .314$.

When cardiorespiratory fitness level was added into the model, the sample size for the WCST was reduced to $N = 63$, with group sizes as follows: COMT Low DA = 22, COMT High DA = 41. Hierarchical regression showed the interaction between COMT x Fitness Level did not significantly explain any variance in performance total errors $\Delta R^2 = .027$, $\Delta F(1, 56) = 2.02$, $p = 161$. However, the interaction did marginally explain 4.8% of the variance in percent perseverative errors, $\Delta R^2 = .048$, $\Delta F(1, 56) = 3.49$, $p = .067$ (Figure 3.12). Simple slopes analysis showed that percent perseverative errors marginally decreased with increasing VO_2 only in the High DA group, $B = -0.92$, $\beta = -1.38$, $t(56) = -3.60$, $p = .002$, 95% CI [-1.54, -0.48], but not the Low DA group, $B = -0.32$, $\beta = -0.04$, $t(56) = -0.07$, $p = .930$, 95% CI [-0.69, 0.77].

Dual task test. The sample size for the dual task test was $N = 68$, with group sizes as follows: COMT Low DA = 24, COMT High DA = 44. ANCOVA showed the Low DA group was significantly faster for both single, $F(1, 68) = 3.99$, $p = .050$, mean difference = -87.08, $p = .040$, 95% CI [-166.76, -3.55], and dual trials, $F(1, 68) = 8.93$, $p = .004$, mean difference = -149.37, $p = .003$, 95% CI [-242.37, -55.75] (Figure A.20a). Further, the Low DA group also showed a significantly lower RT cost (i.e. dual RT – single RT), $F(1, 68) = 5.90$, $p = .018$, mean difference = -62.30, $p = .015$, 95% CI [-111.92, -15.29] (Figure A.20b). There were no differences seen in error rate for either single, $F(1, 68) < 1$, or dual trials, $F(1, 68) = 1.65$, $p = 204$, suggesting there was no speed accuracy tradeoff.

When cardiorespiratory fitness level was added into the model, the sample size for the dual task test was reduced to $N = 63$, with genotypic group sizes as follows: COMT Low DA = 23, COMT High DA = 40. Hierarchical regression showed the interaction between COMT x Fitness Level did not significantly explain any variance in RT or ER for either trial type (Table 3.27).

Dopamine Tables

Table 3.1 Demographics and physical assessment descriptive statistics for DBH 444 G/A (rs1108580)

	G/G		G/A		A/A			Total	
	n = 41 (10 m)		*n* = 107 (42 m)		*n* = 49 (23 m)			*N* = 197 (75 m)	
Demographics	**Mean**	***SD***	**Mean**	***SD***	**Mean**	***SD***	***F (2, 194)***	**Mean**	***SD***
Age	66.59	(5.44)	66.46	(5.94)	66.24	(5.59)	0.04	66.43	(5.73)
Educuation (years)	15.02	(2.44)	16.33	(3.21)	15.63	(3.15)	2.92^{+}	15.88	(3.08)
mMMSE	54.00	(2.79)	54.79	(2.29)	54.67	(2.39)	1.62	54.60	(2.43)

mMMSE = modified mini mental status exam; m = males; + p ≤ .10

χ^2 (2, *N* = 197) = 4.95, *p* = .084

	G/G		G/A		A/A			Total	
	n = 36 (8 m)		*n* = 90 (34 m)		*n* = 37 (16 m)			*N* = 162 (58 m)	
Physical Assessment	**Mean**	***SD***	**Mean**	***SD***	**Mean**	***SD***	***F (2, 159)***	**Mean**	***SD***
Body mass index ($kg \cdot m^{-2}$)	29.89	(3.93)	28.49	(4.51)	28.89	(4.13)	1.36	28.89	(4.32)
VO2 ($mL \cdot kg^{-1} \cdot min^{-1}$)	20.13	(4.93)	21.82	(4.58)	20.94	(4.86)	1.74	21.25	(4.75)

m = males

χ^2 (2, *N* = 163) = 3.94, *p* = .139

Table 3.2 DBH 444 G/A (rs1108580) means and standard deviations

	G/G		G/A		A/A		Total	
	Mean	(***SD***)	**Mean**	(***SD***)	**Mean**	(***SD***)	**Mean**	(***SD***)
Verbal Crystallized Intelligence								
KBIT	109.07	(12.41)	113.21	(9.54)	113.46	(9.87)	112.42	(10.36)
Memory Tests								
Digit Span								
Forward	6.30	(1.24)	6.56	(1.18)	6.53	(1.06)	6.50	(1.16)
Backward	4.73	(1.28)	4.89	(1.38)	5.14	(1.49)	4.92	(1.39)
Spatial Memory								
Load 1 RT (ms)	781.23	(161.18)	811.17	(190.22)	770.91	(175.55)	794.92	(181.02)
Load 2 RT (ms)	898.30	(150.56)	910.53	(194.68)	871.54	(157.68)	897.13	(177.48)
Load 3 RT (ms)	986.53	(160.37)	996.52	(210.71)	957.38	(174.13)	984.52	(192.33)
Load 1 Error Rate	0.10	(0.10)	0.10	(0.07)	0.11	(0.09)	0.10	(0.08)
Load 2 Error Rate	0.15	(0.09)	0.16	(0.11)	0.15	(0.10)	0.16	(0.10)
Load 3 Error Rate	0.20	(0.13)	0.20	(0.11)	0.19	(0.10)	0.20	(0.11)
Executive Control Function Tests								
Task Switch								
Single Condition RT (ms)	777.62	(126.93)	783.16	(114.19)	732.14	(99.01)	770.07	(114.66)
Mix Condition RT (ms)	1170.26	(163.56)	1162.21	(158.93)	1110.74	(131.55)	1151.61	(154.61)
Global RT Cost (ms)	392.64	(122.30)	379.05	(134.06)	378.60	(127.69)	381.54	(129.71)
Repeat Trial RT (ms)	984.17	(137.99)	991.78	(161.54)	926.18	(102.75)	974.87	(147.00)
Switch Trial RT (ms)	1356.35	(226.51)	1332.63	(194.90)	1295.30	(189.64)	1328.36	(199.82)
Local Cost (ms)	372.18	(183.57)	340.84	(164.70)	369.12	(154.31)	353.50	(165.69)
Single Condition Error Rate	0.09	(0.11)	0.05	(0.05)	0.09	(0.12)	0.06	(0.08)
Mix Condition Error Rate	0.16	(0.14)	0.11	(0.09)	0.10	(0.10)	0.12	(0.11)
Global Error Rate Cost	0.07	(0.12)	0.06	(0.09)	0.01	(0.12)	0.05	(0.11)
Repeat Error Rate	0.14	(0.13)	0.09	(0.09)	0.08	(0.10)	0.10	(0.10)
Switch Error Rate	0.19	(0.16)	0.13	(0.10)	0.11	(0.12)	0.14	(0.12)
Local Error Rate Cost	0.05	(0.07)	0.04	(0.06)	0.03	(0.06)	0.04	(0.06)

RT = response time

Table 3.3 Summary of hierarchical regression analysis of the effect of the interaction between DBH 444 G/A x Fitness Level on performance of the spatial memory test

	ΔR^2	ΔF (2, 135)	*p*
Memory Load 1			
Response Time	0.027	2.056	.132
Error Rate	0.001	0.052	.949
Memory Load 2			
Response Time	0.025	1.955	.146
Error Rate	0.020	1.466	.235
Memory Load 3			
Response Time	0.009	0.696	.500
Error Rate	0.003	0.214	.808

Table 3.4 Demographics and physical assessment descriptive statistics for DBH -1021 C/T (rs1611115)

	C/C		**T allele**			**Total**	
	***n* = 139 (46 m)**		***n* = 58 (29 m)**			***N* = 197 (75 m)**	
Demographics	**Mean**	***SD***	**Mean**	***SD***	***F (1, 195)***	**Mean**	***SD***
Age	66.45	(5.69)	66.40	(5.86)	0.00	66.43	(5.73)
Educuation (years)	15.56	(2.84)	16.66	(3.51)	5.27*	15.88	(3.08)
mMMSE	54.23	(2.60)	55.48	(1.69)	11.43*	54.60	(2.43)

*mMMSE = modified mini mental status exam; m = males; *p ≤ .05* χ^2 (1, *N* = 197) = 4.96, *p* = .026*

	C/C		**T allele**			**Total**	
	***n* = 112 (34 m)**		***n* = 50 (24 m)**			***N* = 162 (58 m)**	
Physical Assessment	**Mean**	***SD***	**Mean**	***SD***	***F (1, 160)***	**Mean**	***SD***
Body mass index (kg·m^{-2})	28.81	(4.50)	29.06	(3.88)	0.11	28.89	(4.32)
VO2 (mL·kg^{-1}·min^{-1})	20.71	(4.71)	22.28	(4.58)	3.93	21.19	(4.71)

m = males χ^2 (1, *N*=162) = 4.68, *p* = .030*

Table 3.5 DBH -1021 C/T (rs1611115) means and standard deviations

	C/C		T Carriers		Total	
	Mean	(*SD*)	Mean	(*SD*)	Mean	(*SD*)
Verbal Crystallized Intelligence						
KBIT	111.38	(10.83)	114.84	(8.78)	112.42	(10.36)
Memory Tests						
Digit Span						
Forward	6.42	(1.15)	6.69	(1.19)	6.50	(1.16)
Backward	4.74	(1.22)	5.34	(1.66)	4.92	(1.39)
Spatial Memory						
Load 1 RT (ms)	817.80	(179.86)	743.23	(174.41)	794.92	(181.02)
Load 2 RT (ms)	920.02	(177.48)	845.42	(167.80)	897.13	(177.48)
Load 3 RT (ms)	1006.58	(194.58)	934.69	(179.03)	984.52	(192.33)
Load 1 Error Rate	0.11	(0.09)	0.09	(0.07)	0.10	(0.08)
Load 2 Error Rate	0.12	(0.10)	0.17	(0.10)	0.16	(0.10)
Load 3 Error Rate	0.20	(0.11)	0.19	(0.11)	0.20	(0.11)
Executive Control Function Tests						
Flanker Test						
Congruent RT (ms)	595.12	(89.82)	562.08	(93.12)	585.40	(91.80)
Incongruent RT (ms)	680.25	(111.45)	639.15	(109.59)	668.17	(112.20)
RT Cost (ms)	85.14	(56.25)	77.07	(57.50)	82.76	(56.58)
Congruent Error Rate	0.02	(0.03)	0.01	(0.03)	0.01	(0.03)
Incongruent Error Rate	0.06	(0.08)	0.04	(0.07)	0.06	(0.08)
Error Rate Cost	0.05	(0.07)	0.03	(0.06)	0.04	(0.07)
WCST						
Total Errors	43.98	(23.31)	37.96	(23.82)	42.30	(23.54)
Percent Perseverative Errors	17.39	(8.63)	17.45	(11.03)	17.41	(9.34)
Task Switch						
Single Condition RT (ms)	777.49	(116.78)	754.21	(109.45)	770.07	(114.66)
Mix Condition RT (ms)	1179.02	(149.15)	1092.96	(151.10)	1151.61	(154.61)
Global RT Cost (ms)	401.54	(133.45)	338.75	(110.92)	381.54	(129.71)
Repeat Trial RT (ms)	994.37	(152.73)	933.12	(125.38)	974.87	(147.00)
Switch Trial RT (ms)	1363.67	(189.69)	1252.80	(201.83)	1328.36	(199.82)
Local Cost (ms)	369.30	(172.15)	319.68	(146.90)	353.50	(165.69)
Single Condition Error Rate	0.06	(0.08)	0.07	(0.09)	0.06	(0.08)
Mix Condition Error Rate	0.13	(0.11)	0.09	(0.10)	0.12	(0.11)
Global Error Rate Cost	0.07	(0.10)	0.02	(0.11)	0.05	(0.11)
Repeat Error Rate	0.11	(0.10)	0.07	(0.10)	0.10	(0.10)
Switch Error Rate	0.15	(0.12)	0.11	(0.11)	0.14	(0.12)
Local Error Rate Cost	0.04	(0.06)	0.04	(0.05)	0.04	(0.06)
Dual Task						
Single RT (ms)	1245.29	(171.16)	1183.30	(140.13)	1225.67	(164.10)
Dual RT (ms)	2036.62	(188.35)	1969.10	(214.15)	2015.25	(198.70)
RT Cost (ms)	791.33	(124.43)	785.80	(134.69)	789.58	(127.36)
Single Error Rate	0.06	(0.08)	0.06	(0.08)	0.06	(0.08)
Dual Error Rate	0.15	(0.14)	0.12	(0.13)	0.14	(0.14)
Error Rate Cost (ms)	0.09	(0.10)	0.06	(0.08)	0.08	(0.10)

RT = response time

Table 3.6 Summary of hierarchical regression analysis of the effect of the interaction between DBH -1021 C/T x Fitness Level on performance of the spatial memory test

	ΔR^2	$\Delta F(1, 136)$	p
Memory Load 1			
Response Time	0.002	0.281	.597
Error Rate	0.000	0.001	.973
Memory Load 2			
Response Time	0.004	0.705	.403
Error Rate	0.003	0.386	.535
Memory Load 3			
Response Time	0.009	1.373	.243
Error Rate	0.001	0.239	.625

Table 3.7 Summary of hierarchical regression analysis of the effect of the interaction between DBH -1021 C/T x Fitness Level on performance of the Flanker test

	ΔR^2	$\Delta F(1, 145)$	p
Congruent Trial Type			
Response Time	0.004	0.652	.421
Error Rate^	0.028	4.239	.041
Incongruent Trial Type			
Response Time	0.004	0.683	.410
Error Rate	0.002	0.291	.590

^Full model was not significant, $F(7, 145) = 1.098$, $p = .368$

Table 3.8 Summary of hierarchical regression analysis of the effect of the interaction between DBH -1021 C/T x Fitness Level on performance of the task switch test

	ΔR^2	$\Delta F(1, 123)$	p
Single Condition			
Response Time	0.004	0.479	.490
Error Rate	0.016	2.145	.146
Mix Condition			
Response Time	0.000	0.002	.962
Error Rate	0.002	0.229	.633
Repeat Trial Type			
Response Time	0.005	0.665	.417
Error Rate	0.001	0.083	.773
Switch Trial Type			
Response Time	0.003	0.402	.527
Error Rate	0.003	0.360	.550

Table 3.9 Summary of hierarchical regression analysis of the effect of the interaction between DBH -1021 C/T x Fitness Level on performance of the dual task test

	ΔR^2	$\Delta F(1, 143)$	p
Single Trial Type			
Response Time	0.004	0.739	.391
Error Rate	0.002	0.300	.584
Dual Trial Type			
Response Time	0.012	2.137	.146
Error Rate	0.016	2.720	.101

Table 3.10 Demographics and physical assessment descriptive statistics for DBH Combined

	DBH Low DA	DBH High DA		Total
	n = 33 (10 m)	*n* = 17 (10 m)		*N* = 50 (20 m)
Demographics	**Mean *SD***	**Mean *SD***	***F* (1, 48)**	**Mean *SD***
Age	66.09 (5.41)	65.12 (5.20)	0.37	65.76 (5.30)
Educuation (years)	15.03 (2.48)	16.41 (3.74)	2.44	15.50 (3.01)
mMMSE	53.73 (2.98)	55.76 (1.30)	7.72*	54.42 (2.70)

MMSE = modified mini mental status exam; m = males; * $p \leq .05$; + $p \leq .10$ $^{+}\chi^2$ (1, *N*=50) = 3.80, *p* = .051

	DBH Low DA	DBH High DA		Total
	n = 29 (8 m)	*n* = 13 (7 m)		*N* = 42 (15 m)
Physical Assessment	**Mean *SD***	**Mean *SD***	***F* (1, 66)**	**Mean *SD***
Body mass index ($kg \cdot m^{-2}$)	30.48 (3.80)	30.31 (4.09)	0.02	30.43 (3.85)
VO2 ($mL \cdot kg^{-1} \cdot min^{-1}$)	20.26 (5.38)	20.78 (5.82)	0.08	20.42 (5.46)

m = males χ^2 (1, *N*=42) = 2.70, *p* = .101

Table 3.11 DBH SNPs combined means and standard deviations

	DBH Low DA (G/G + C/C)		DBH High DA (A/A + T Allele)		Total	
	Mean	(*SD*)	Mean	(*SD*)	Mean	(*SD*)
Verbal Crystallized Intelligence						
KBIT	107.47	(13.01)	117.94	(9.27)	111.10	(12.78)
Memory Tests						
Digit Span						
Forward	6.34	(1.34)	6.88	(1.22)	6.53	(1.31)
Backward	4.78	(1.41)	6.06	(1.89)	5.22	(1.69)
Spatial Memory						
Load 1 RT (ms)	795.29	(160.55)	694.81	(147.10)	758.75	(161.63)
Load 2 RT (ms)	905.54	(152.20)	786.63	(139.89)	862.30	(157.23)
Load 3 RT (ms)	1006.95	(166.95)	890.47	(146.75)	964.59	(168.00)
Load 1 Error Rate	0.11	(0.11)	0.10	(0.08)	0.11	(0.10)
Load 2 Error Rate	0.14	(0.09)	0.15	(0.11)	0.15	(0.10)
Load 3 Error Rate	0.19	(0.12)	0.18	(0.11)	0.19	(0.12)
Executive Control Function Tests						
Flanker Test						
Congruent RT (ms)	614.97	(99.19)	538.37	(92.65)	590.52	(102.69)
Incongruent RT (ms)	693.39	(117.34)	613.80	(99.73)	667.99	(117.10)
RT Cost (ms)	78.42	(58.12)	75.43	(41.81)	77.47	(53.01)
Congruent Error Rate	0.02	(0.03)	0.01	(0.02)	0.01	(0.03)
Incongruent Error Rate	0.08	(0.10)	0.03	(0.04)	0.06	(0.09)
Error Rate Cost	0.06	(0.10)	0.03	(0.04)	0.05	(0.09)
Task Switch						
Single Condition RT (ms)	788.82	(130.05)	701.12	(85.67)	753.32	(121.09)
Mix Condition RT (ms)	1190.62	(161.05)	1050.21	(128.35)	113.79	(162.72)
Global RT Cost (ms)	401.80	(129.84)	349.09	(86.65)	380.46	(116.12)
Repeat Trial RT (ms)	1001.54	(134.64)	884.74	(87.33)	954.26	(130.21)
Switch Trial RT (ms)	1379.70	(229.61)	1215.68	(181.98)	1313.31	(224.56)
Local Cost (ms)	378.16	(194.80)	330.94	(124.85)	359.05	(169.84)
Single Condition Error Rate	0.08	(0.11)	0.10	(0.13)	0.09	(0.12)
Mix Condition Error Rate	0.16	(0.14)	0.07	(0.09)	0.12	(0.13)
Global Error Rate Cost	0.08	(0.12)	-0.03	(0.12)	0.03	(0.13)
Repeat Error Rate	0.13	(0.13)	0.06	(0.08)	0.11	(0.12)
Switch Error Rate	0.18	(0.16)	0.08	(0.10)	0.14	(0.14)
Local Error Rate Cost	0.05	(0.07)	0.02	(0.05)	0.03	(0.07)

RT = response time

Table 3.12 Summary of hierarchical regression analysis of the effect of the interaction between DBH Combined x Fitness Level on performance of the spatial memory test

	ΔR^2	$\Delta F(1, 28)$	p
Memory Load 1			
Response Time	0.010	0.341	.564
Error Rate	0.002	0.09	.766
Memory Load 2			
Response Time	0.021	0.805	.377
Error Rate	0.009	0.332	.569
Memory Load 3			
Response Time	0.007	0.26	.614
Error Rate	0.002	0.074	.787

Table 3.13 Summary of hierarchical regression analysis of the effect of the interaction between DBH Combined x Fitness Level on performance of the Flanker test

	ΔR^2	$\Delta F(1, 29)$	p
Congruent Trial Type			
Response Time	0.007	0.246	.623
Error Rate^	0.082	3.29	.079
Incongruent Trial Type			
Response Time	0.021	0.805	.376
Error Rate	0.010	0.385	.539

^Full model was not significant, F (7, 32) = 1.18, p = .341

Table 3.14 Summary of hierarchical regression analysis of the effect of the interaction between DBH Combined x Fitness Level on performance of the task switch test

	ΔR^2	$\Delta F(1, 27)$	p
Single Condition			
Response Time	0.000	0.000	.996
Error Rate	0.000	0.001	.974
Mix Condition			
Response Time	0.014	0.714	.406
Error Rate	0.023	1.290	.266
Repeat Trial Type			
Response Time	0.019	0.993	.328
Error Rate	0.005	0.234	.633
Switch Trial Type			
Response Time	0.008	0.377	.544
Error Rate	0.044	2.770	.108

Table 3.15 Demographics and physical assessment descriptive statistics for COMT Val158Met (rs4680)

	Val/Val (G/G)		Val/Met (G/A)		Met/Met (A/A)			Total	
	n = 32 (14 m)		*n* = 107 (35 m)		*n* = 57 (26 m)			*N* = 196 (75 m)	
Demographics	**Mean**	***SD***	**Mean**	***SD***	**Mean**	***SD***	***F (2, 193)***	**Mean**	***SD***
Age	67.75	(5.25)	66.11	(5.81)	66.21	(5.84)	1.05	66.41	(5.73)
Educuation (years)	15.31	(2.81)	16.02	(3.32)	15.95	(2.80)	0.66	15.88	(3.09)
mMMSE	54.63	(2.43)	54.64	(2.23)	54.46	(2.80)	0.12	54.59	(2.43)

mMMSE = modified mini mental status exam; m = males χ^2 (2, N = 196) = 3.11, p = .211

	Val/Val (G/G)		Val/Met (G/A)		Met/Met (A/A)			Total	
	n = 28 (11 m)		*n* = 87 (28 m)		*n* = 46 (19 m)			*N* = 161 (58 m)	
Physical Assessment	**Mean**	***SD***	**Mean**	***SD***	**Mean**	***SD***	***F (2, 158)***	**Mean**	***SD***
Body mass index ($kg \cdot m^{-2}$)	28.96	(4.36)	29.08	(4.46)	28.57	(4.09)	0.22	28.91	(4.32)
VO2 ($mL \cdot kg^{-1} \cdot min^{-1}$)	20.10	(3.70)	21.20	(4.81)	21.86	(5.08)	1.21	21.19	(4.73)

m = males χ^2 (2, N =161) = 1.24 p = .537

Table 3.16 COMT G/A (rs4680) means and standard deviations

	Val/Val (G/G)	Val/Met (G/A)	Met/Met (A/A)	Total
	Mean (*SD*)	Mean (*SD*)	Mean (*SD*)	Mean (*SD*)
Verbal Crystallized Intelligence				
KBIT	113.42 (11.02)	112.03 (10.41)	112.35 (9.99)	112.35 (10.34)
Memory Tests				
Digit Span				
Forward	6.23 (1.06)	6.48 (1.17)	6.67 (1.19)	6.49 (1.16)
Backward	5.23 (1.31)	4.89 (1.40)	4.79 (1.40)	4.91 (1.39)
Spatial Memory				
Load 1 RT (ms)	719.20 (169.98)	794.22 (173.50)	830.38 (185.81)	792.76 (179.24)
Load 2 RT (ms)	832.95 (178.96)	896.84 (165.09)	929.28 (192.65)	896.07 (177.42)
Load 3 RT (ms)	910.75 (200.13)	974.53 (174.87)	1039.34 (205.89)	983.21 (192.10)
Load 1 Error Rate	0.12 (0.08)	0.10 (0.08)	0.10 (0.10)	0.10 (0.08)
Load 2 Error Rate	0.15 (0.10)	0.15 (0.09)	0.17 (0.11)	0.16 (0.10)
Load 3 Error Rate	0.20 (0.13)	0.19 (0.10)	0.21 (0.12)	0.20 (0.11)
Executive Control Function Tests				
WCST				
Total Errors	40.54 (21.54)	43.19 (23.27)	42.13 (25.18)	42.46 (23.51)
Percent Perseverative Errors	15.71 (6.00)	17.69 (9.57)	17.93 (10.29)	17.46 (9.33)
Flanker Test				
Congruent RT (ms)	581.07 (115.38)	576.79 (77.98)	603.53 (100.68)	585.38 (92.05)
Incongruent RT (ms)	659.37 (128.38)	656.43 (91.46)	694.98 (134.07)	668.30 (112.48)
RT Cost (ms)	78.30 (50.51)	79.64 (49.06)	91.45 (71.27)	82.92 (56.70)
Congruent Error Rate	0.02 (0.03)	0.01 (0.03)	0.02 (0.03)	0.01 (0.03)
Incongruent Error Rate	0.10 (0.12)	0.05 (0.06)	0.06 (0.07)	0.06 (0.08)
Error Rate Cost	0.07 (0.11)	0.03 (0.05)	0.04 (0.06)	0.04 (0.07)
Dual Task				
Single RT (ms)	1156.99 (148.66)	1247.16 (162.36)	1229.46 (169.02)	1226.35 (164.40)
Dual RT (ms)	1924.52 (197.53)	2029.93 (187.88)	2041.56 (209.41)	2015.28 (199.34)
RT Cost (ms)	767.53 (104.87)	782.77 (135.48)	812.09 (123.71)	788.93 (127.49)
Single Error Rate	0.05 (0.06)	0.07 (0.09)	0.05 (0.07)	0.06 (0.08)
Dual Error Rate	0.10 (0.12)	0.17 (0.14)	0.12 (0.14)	0.14 (0.14)
Error Rate Cost (ms)	0.05 (0.07)	0.09 (0.09)	0.08 (0.12)	0.08 (0.10)

RT = response time

Table 3.17 Summary of hierarchical regression analysis of the effect of the interaction between COMT Val158Met x Fitness Level on performance of the spatial memory test

	ΔR^2	$\Delta F(2, 134)$	p
Memory Load 1			
Response Time	0.007	0.571	.566
Error Rate	0.021	1.575	.211
Memory Load 2			
Response Time	0.007	0.597	.552
Error Rate	0.004	0.288	.750
Memory Load 3			
Response Time	0.013	1.010	.367
Error Rate	0.015	1.230	.296

Table 3.18 Summary of hierarchical regression analysis of the effect of the interaction between COMT Val158Met x Fitness Level on performance of the Flanker test

	ΔR^2	$\Delta F(2, 143)$	p
Congruent Trial Type			
Response Time	0.035	2.899	.058
Error Rate	0.008	0.593	.554
Incongruent Trial Type			
Response Time	0.016	1.237	.293
Error Rate	0.011	0.809	.447

Table 3.19 Summary of simple slopes regression analysis of the relationship between cardiorespiratory fitness level on response time for congruent trials of the Flanker test for each genotypic group of the COMT Val158Met SNP

					95% Confidence Interval	
Congruent Trial RT	B	β	$t(143)$	p	Lower	Upper
Val/Val	-8.658	-0.166	-1.839	.114	-21.665	1.325
Val/Met	1.578	0.062	0.681	.450	-2.418	5.612
Met/Met	3.955	0.120	1.376	.163	-1.81	9.75

Table 3.20 Demographics and physical assessment descriptive statistics for COMT C/G (rs4818)

	G Carriers	C/C		Total
	n = 116 (45 m)	*n* = 80 (30 m)		*N* = 196 (75 m)
Demographics	**Mean *SD***	**Mean *SD***	***F (1, 194)***	**Mean *SD***
Age	66.41 (5.76)	66.40 (5.73)	0.00	66.41 (5.73)
Educuation (years)	15.87 (3.27)	15.90 (2.84)	0.00	15.88 (3.09)
mMMSE	54.68 (2.35)	54.45 (2.56)	0.43	54.59 (2.43)

MMSE = modified mini mental status exam; m = males

χ^2 (1, *N*=196) = 0.89, *p* = .855

	G Carriers	C/C		Total
	n = 95 (36 m)	*n* = 66 (22 m)		*N* = 161 (58 m)
Physical Assessment	**Mean *SD***	**Mean *SD***	***F (1, 159)***	**Mean *SD***
Body mass index ($kg \cdot m^{-2}$)	29.34 (4.29)	28.30 (4.32)	2.25	28.91 (4.32)
VO2 ($mL \cdot kg^{-1} \cdot min^{-1}$)	21.22 (4.51)	21.15 (5.06)	0.01	21.19 (4.73)

m = males

χ^2 (1, *N*=161) = 0.35 *p* = .553

Table 3.21 COMT C/G (rs4818) means and standard deviations

	G Carriers	C/C	Total
	Mean (*SD*)	Mean (*SD*)	Mean (*SD*)
Verbal Crystallized Intelligence			
KBIT	112.35 (10.62)	112.35 (10.01)	112.35 (10.34)
Memory Tests			
Digit Span			
Forward	6.44 (1.19)	6.56 (1.11)	6.49 (1.16)
Backward	5.05 (1.43)	4.71 (1.31)	4.91 (1.39)
Spatial Memory			
Load 1 RT (ms)	770.81 (172.70)	825.68 (185.01)	792.76 (179.24)
Load 2 RT (ms)	876.71 (167.82)	925.11 (188.44)	896.07 (177.42)
Load 3 RT (ms)	954.42 (178.41)	1026.41 (204.76)	983.21 (192.10)
Load 1 Error Rate	0.10 (0.08)	0.10 (0.09)	0.10 (0.08)
Load 2 Error Rate	0.16 (0.09)	0.16 (0.11)	0.16 (0.10)
Load 3 Error Rate	0.19 (0.11)	0.21 (0.12)	0.20 (0.11)
Executive Control Function Tests			
Flanker Test			
Congruent RT (ms)	572.87 (91.19)	603.50 (90.85)	585.38 (92.05)
Incongruent RT (ms)	655.30 (107.49)	687.12 (117.53)	668.30 (112.48)
RT Cost (ms)	82.43 (51.98)	83.62 (63.26)	82.92 (56.70)
Congruent Error Rate	0.02 (0.03)	0.01 (0.02)	0.01 (0.03)
Incongruent Error Rate	0.06 (0.08)	0.06 (0.08)	0.06 (0.08)
Error Rate Cost	0.04 (0.07)	0.04 (0.07)	0.04 (0.07)
WCST			
Total Errors	41.90 (22.60)	43.24 (24.84)	42.46 (23.51)
Percent Perseverative Errors	17.14 (8.88)	17.89 (9.97)	17.46 (9.33)
Dual Task Test			
Single RT (ms)	1218.45 (165.63)	1237.26 (163.33)	1226.35 (164.40)
Dual RT (ms)	1987.73 (191.87)	2053.27 (204.61)	2015.28 (199.34)
RT Cost (ms)	769.28 (126.15)	816.01 (125.27)	788.93 (127.49)
Single Error Rate	0.06 (0.07)	0.06 (0.09)	0.06 (0.08)
Dual Error Rate	0.14 (0.13)	0.15 (0.16)	0.14 (0.14)
Error Rate Cost	0.08 (0.08)	0.09 (0.12)	0.08 (0.10)

RT = response time

Table 3.22 Summary of hierarchical regression analysis of the effect of the interaction between COMT C/G x Fitness Level on performance of the spatial memory test

	ΔR^2	$\Delta F(2, 134)$	p
Memory Load 1			
Response Time	0.001	0.040	.961
Error Rate	0.002	0.148	.863
Memory Load 2			
Response Time	0.001	0.049	.952
Error Rate	0.015	1.080	.342
Memory Load 3			
Response Time	0.002	0.184	.832
Error Rate	0.002	0.176	.839

Table 3.23 Summary of hierarchical regression analysis of the effect of the interaction between COMT C/G x Fitness Level on performance of the Flanker test

	ΔR^2	$\Delta F(2, 143)$	p
Congruent Trial Type			
Response Time	0.000	0.024	.976
Error Rate	0.001	0.071	.932
Incongruent Trial Type			
Response Time	0.003	0.264	.768
Error Rate	0.006	0.422	.657

Table 3.24 Demographics and physical assessment descriptive statistics for COMT Combined

	COMT Low DA		COMT High DA			Total	
	n = 29 (14 m)		*n* = 53 (23 m)			*N* = 82 (37 m)	
Demographics	**Mean**	***SD***	**Mean**	***SD***	***F* (1, 80)**	**Mean**	***SD***
Age	67.86	(5.47)	66.28	(5.73)	1.47	66.84	(5.66)
Educuation (years)	15.10	(2.76)	15.75	(2.72)	1.06	15.52	(2.74)
mMMSE	54.93	(2.31)	54.57	(2.64)	0.39	54.70	(2.52)

MMSE = modified mini mental status exam; m = males

χ^2 (1, *N*=82) = 0.18, *p* = .671

	COMT Low DA		COMT High DA			Total	
	n = 25 (11 m)		*n* = 43 (16 m)			*N* = 68 (27 m)	
Physical Assessment	**Mean**	***SD***	**Mean**	***SD***	***F* (1, 66)**	**Mean**	***SD***
Body mass index ($kg \cdot m^{-2}$)	28.40	(4.17)	28.72	(4.13)	0.10	28.60	(4.12)
VO2 ($mL \cdot kg^{-1} \cdot min^{-1}$)	20.55	(3.62)	21.50	(4.98)	0.07	21.15	(4.52)

m = males

χ^2 (1, *N*=68) = 0.31 *p* = .581

Table 3.25 COMT SNPs combined means and standard deviations

	Low DA (Val/Val + G Allele)		High DA (Met/Met + C/C)		Total	
	Mean	(*SD*)	Mean	(*SD*)	Mean	(*SD*)
Verbal Crystallized Intelligence						
KBIT	113.29	(11.33)	112.43	(9.89)	112.73	(10.35)
Memory Tests						
Digit Span						
Forward	6.29	(1.08)	6.64	(1.16)	6.52	(1.14)
Backward	5.32	(1.34)	4.79	(1.39)	4.98	(1.39)
Spatial Memory						
Load 1 RT (ms)	726.76	(168.35)	831.13	(189.41)	794.05	(187.92)
Load 2 RT (ms)	840.50	(177.76)	928.28	(196.48)	897.09	(193.52)
Load 3 RT (ms)	919.33	(198.62)	1037.57	(209.70)	995.57	(212.29)
Load 1 Error Rate	0.12	(0.08)	0.11	(0.10)	0.11	(0.09)
Load 2 Error Rate	0.15	(0.10)	0.17	(0.12)	0.16	(0.11)
Load 3 Error Rate	0.20	(0.13)	0.21	(0.12)	0.21	(0.12)
Executive Control Function Tests						
WCST						
Total Errors	40.31	(21.99)	40.35	(24.48)	40.34	(23.53)
Percent Perseverative Errors	15.88	(6.16)	16.98	(9.57)	16.61	(8.54)
Dual Task						
Single RT (ms)	1155.70	(157.27)	1231.97	(173.08)	1205.05	(170.48)
Dual RT (ms)	1918.70	(202.87)	2044.31	(211.27)	1999.97	(215.48)
RT Cost (ms)	763.00	(102.16)	812.33	(125.56)	794.92	(119.43)
Single Error Rate	0.05	(0.06)	0.05	(0.07)	0.05	(0.07)
Dual Error Rate	0.10	(0.13)	0.12	(0.15)	0.11	(0.14)
Error Rate Cost (ms)	0.05	(0.08)	0.07	(0.12)	0.07	(0.11)

RT = response time

Table 3.26 Summary of hierarchical regression analysis of the effect of the interaction between COMT Combined x Fitness Level on performance of the spatial memory test

	ΔR^2	$\Delta F(2, 134)$	p
Memory Load 1			
Response Time	0.025	1.984	.164
Error Rate	0.008	0.522	.473
Memory Load 2			
Response Time	0.021	1.740	.192
Error Rate	0.000	0.000	.998
Memory Load 3			
Response Time	0.029	2.396	.127
Error Rate	0.001	0.046	.832

Table 3.27 Summary of hierarchical regression analysis of the effect of the interaction between COMT Combined x Fitness Level on performance of the dual task test

	ΔR^2	$\Delta F(1, 56)$	p
Single Trial Type			
Response Time	0.003	0.186	.668
Error Rate	0.000	0.017	.897
Dual Trial Type			
Response Time	0.000	0.013	.910
Error Rate	0.016	0.994	.323

Dopamine Figures

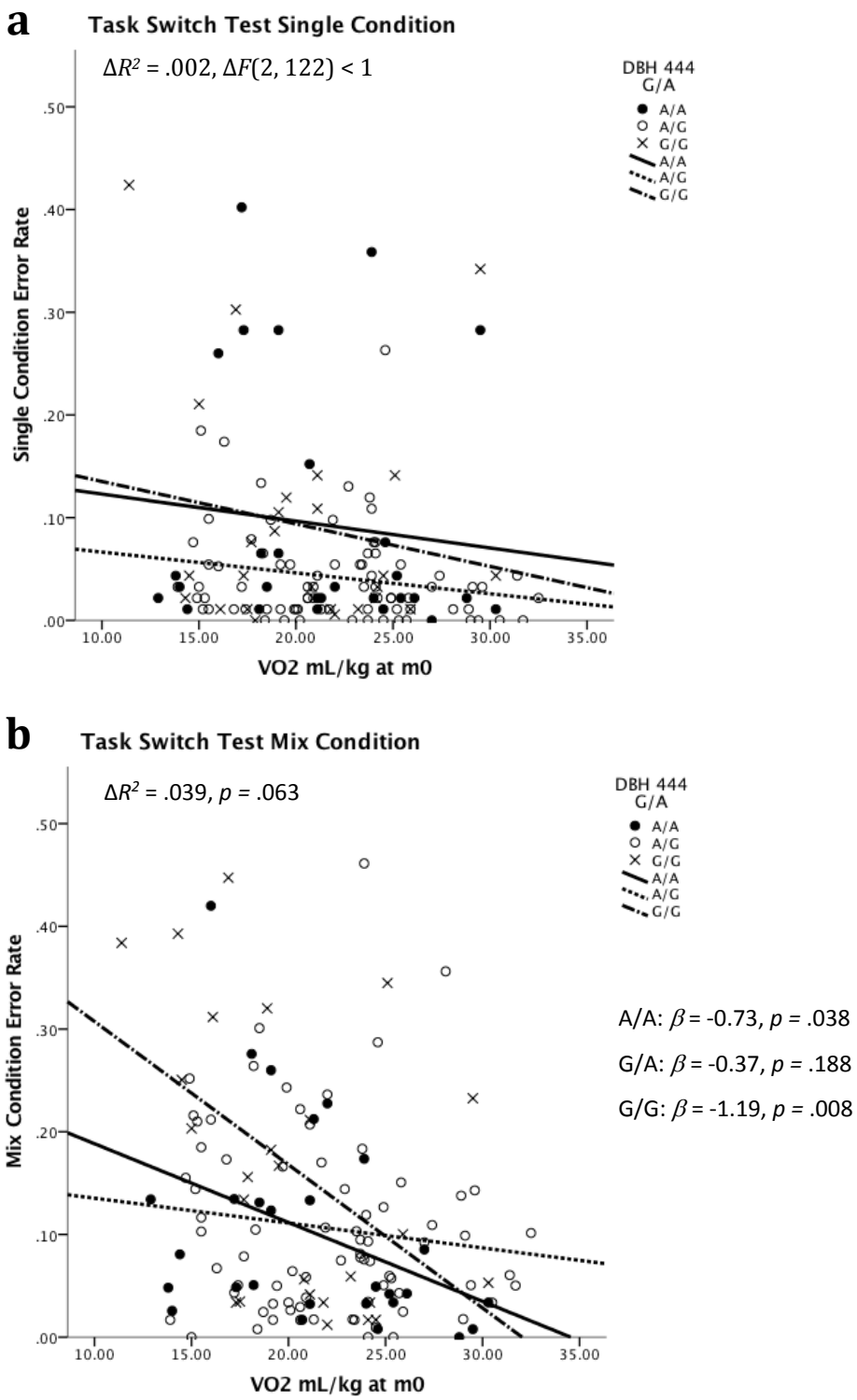


Figure 3.1 Interaction effect between DBH 44 G/A and cardiorespiratory fitness level (i.e. VO_2) on error rate (ER) during the two conditions (i.e. single and mix) of the task switch test.
a) Single condition: There was no significant interaction effect on ER for the single condition, $\Delta R^2 = .002$, $\Delta F(2, 122) < 1$ **b)** Mix condition: The interaction did marginally explain 3.9% of the variance in ER for the mix

condition, ΔR^2 = .039, $\Delta F(2, 122)$ = 2.82, p = .063. Simple slopes: A/A group, B = -0.009, β = -0.73, $t(122)$ = -2.02, p = .038, 95% CI [-0.018, -0.001]; G/A group, B = -0.003, β = -0.37, $t(122)$ = -1.17, p = .188, 95% CI [-0.008, 0.002]; G/G group, B = -0.015, β = -1.19, $t(122)$ = -3.25, p = .008, 95% CI [-0.027, -0.003]

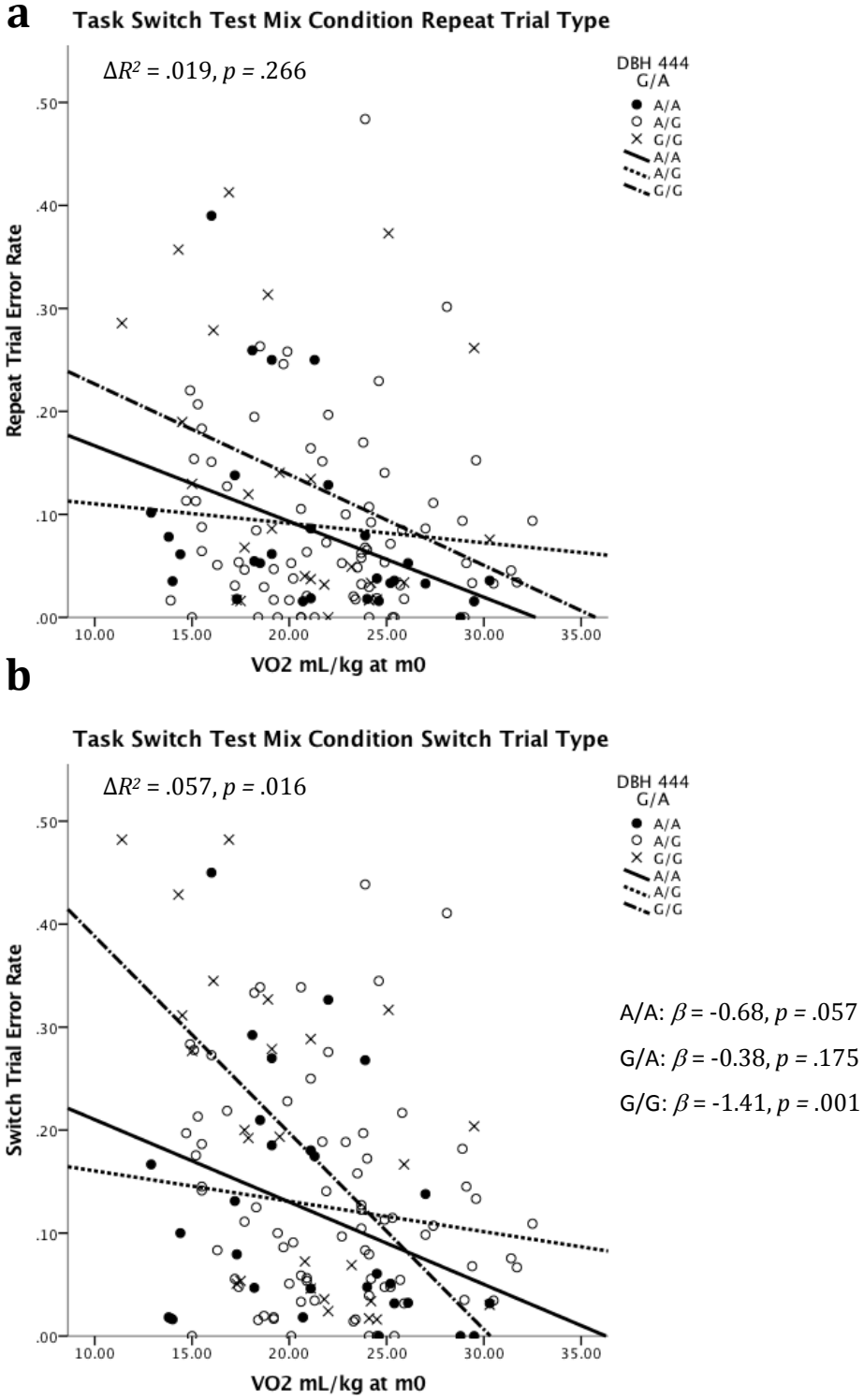


Figure 3.2 Interaction effect between DBH 444 G/A and cardiorespiratory fitness level (i.e. VO_2) on error rate (ER) during the two trial types (i.e. repeat and switch) of the mix condition of the task switch test.

a) Repeat trials: There was no significant interaction effect on ER for repeat trials, $\Delta R^2 = .019$, $\Delta F(2, 122) = 1.34$, $p = .266$ **b)** Switch trials: The interaction did marginally explain 5.7 % of the variance in ER for switch trials, $\Delta R^2 = .057$, $\Delta F(2, 122) = 4.308$, $p = .016$. Simple slopes: A/A group, $B = -0.009$, $\beta = -0.68$, $t(122) = -1.91$, $p = .057$, 95% CI[-0.020, 0.000]; G/A group, $B = -0.004$, $\beta = -0.38$, $t(122) = -1.21$, $p = .175$, 95% CI[-0.009, 0.002]; G/G group, $B = -0.020$, $\beta = -1.41$, $t(122) = -3.93$, $p = .001$, 95% CI[-0.032, -0.009]

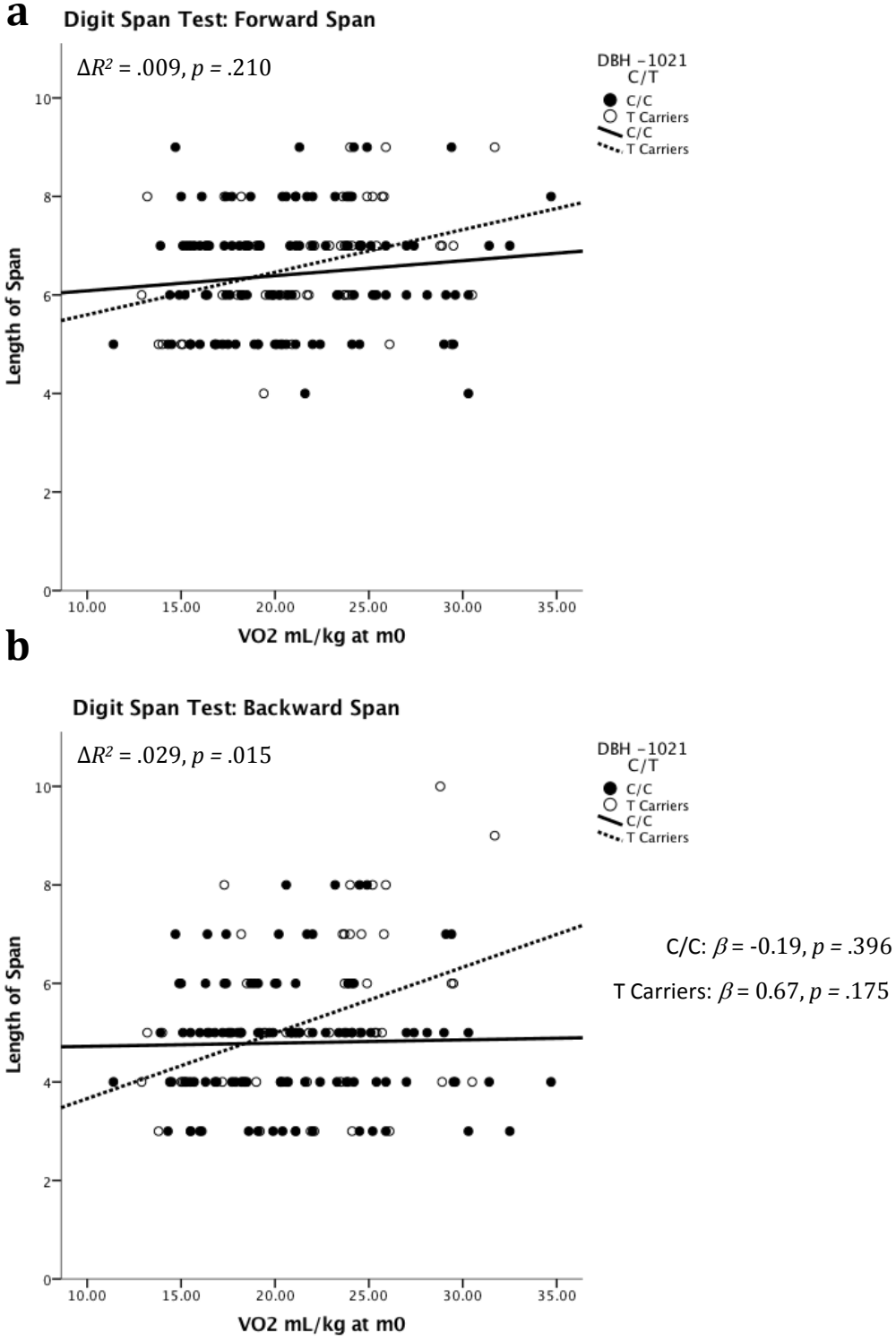


Figure 3.3 Interaction effect between DBH -1021 C/T and cardiorespiratory fitness level (i.e. VO_2) on length of span for each version of the digit span test, forward and backward.
a) Forward span: There was no significant interaction effect on the length of span, $\Delta R^2 = .009$, $\Delta F(1, 153) = 1.59$, $p = .210$ **b)** Backward span: The interaction did significantly explain 2.9% of the variance in backward span, $\Delta R^2 = .029$, $\Delta F(1, 153) = 6.00$, $p = .015$. Simple slopes: C/C Homozygotes, $B = -.026$, $\beta = -0.19$, $t(153) = -0.86$, $p = .396$, 95% CI [-0.080, 0.039]; T Carriers, $B = 0.089$, $\beta = 0.67$, $t(153) = 2.00$, $p = .103$, 95% CI [-0.013,

0.201. The slopes were significantly different from each other, $B = 0.115$, $\beta = .862$, $t(153) = 2.45$, $p = .032$, 95% CI [0.012, 0.221]

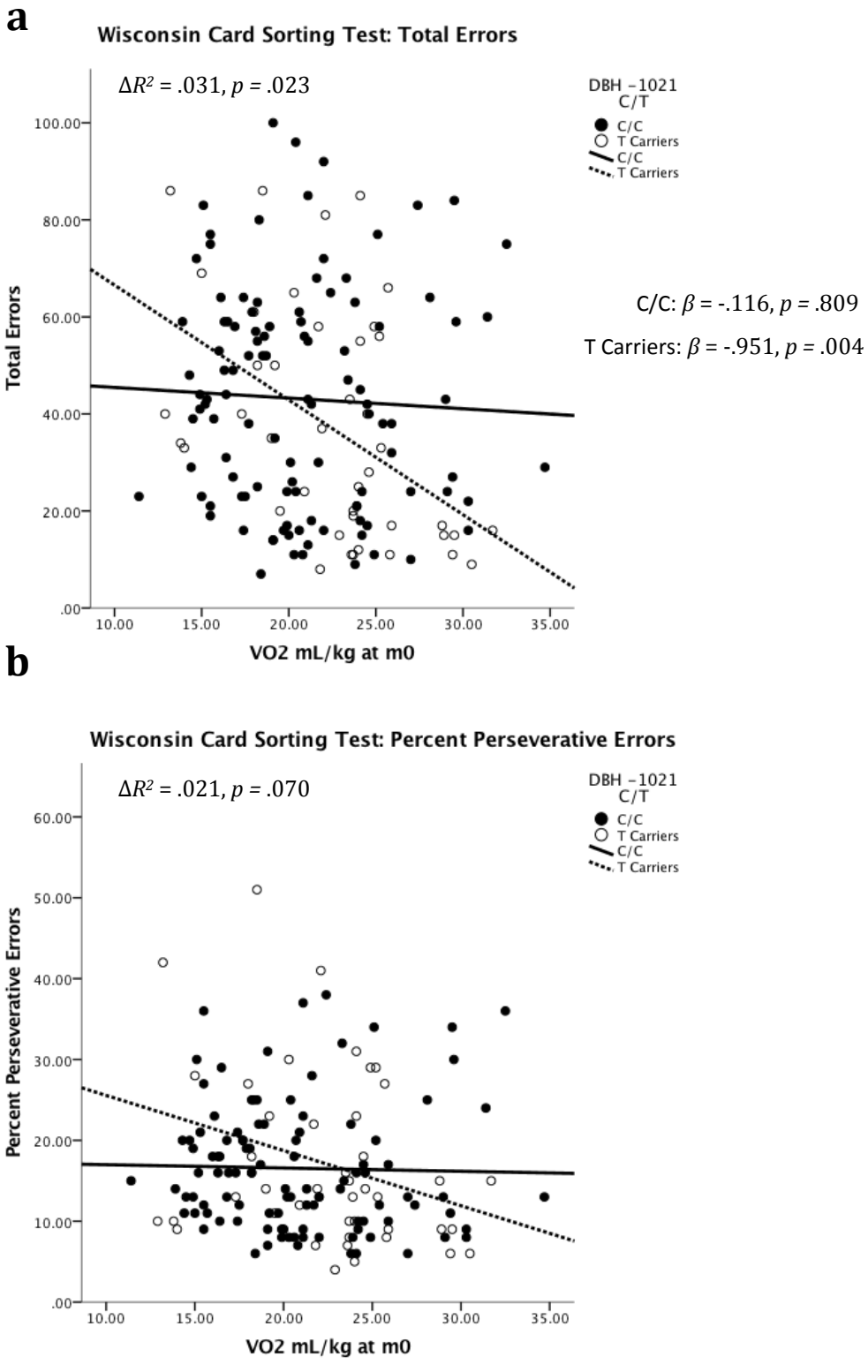


Figure 3.4 Interaction effect between DBH -1021 C/T and cardiorespiratory fitness level (i.e. VO_2) on performance of the Wisconsin Card Sorting Test (WCST) measured by total errors and percent perseverative

errors. **a)** Total errors: The interaction significantly explained 3.1% of the variance in total errors, $\Delta R^2 = .031$, $\Delta F(1, 142) = 5.28$, $p = .023$. Simple slopes: T Carriers, $B = -2.10$, $\beta = -.951$, $t(142) = -2.58$, $p = .004$, 95% CI [-3.57, -0.64]; C/C Homozygotes, $B = -0.13$, $\beta = -.116$, $t(142) = -0.24$, $p = .809$, 95% CI [-1.21, 0.84] **b)** Percent perseverative errors: The interaction marginally explained 2.1% of the variance in percent perseverative errors, $\Delta R^2 = .021$, $\Delta F(1, 142) = 3.33$, $p = .070$. Simple slopes: T Carriers, $B = -0.65$, $\beta = -.780$, $t(142) = -2.05$, $p = .109$, 95% CI [-1.53, 0.11]; C/C Homozygotes, $B = -0.04$, $\beta = -.009$, $t(142) = -0.20$, $p = .851$, 95% CI [-0.47, 0.36]

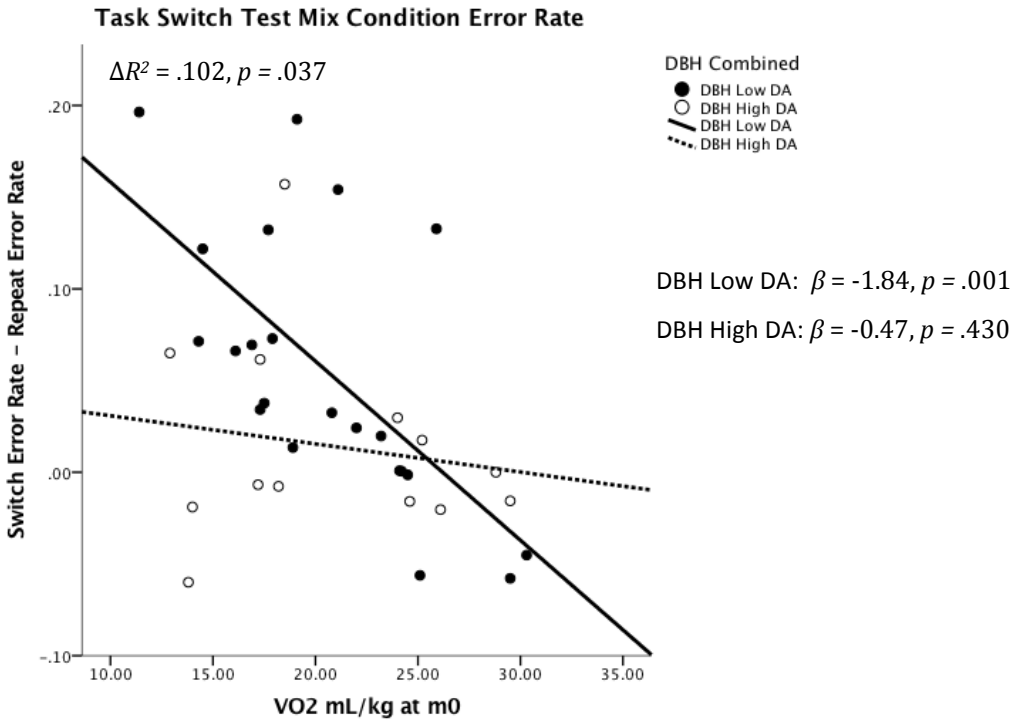


Figure 3.5 Interaction effect between DBH Combined SNPs and cardiorespiratory fitness level (i.e. VO_2) on local error rate cost for the two trial types of the mix condition of the task switch test. The interaction did significantly explain 10.2% of the variance, $\Delta R^2 = .102$, $\Delta F(1, 27) = 4.83$, $p = .037$. Simple slopes: DBH low DA group, $B = -0.012$, $\beta = -1.84$, $t(27) = -3.57$, $p = .001$, 95% CI [-.019, -.005]; DBH High DA group, $B = -0.003$, $\beta = -0.47$, $t(27) = -0.80$, $p = .430$, 95% CI [-.011, .005]

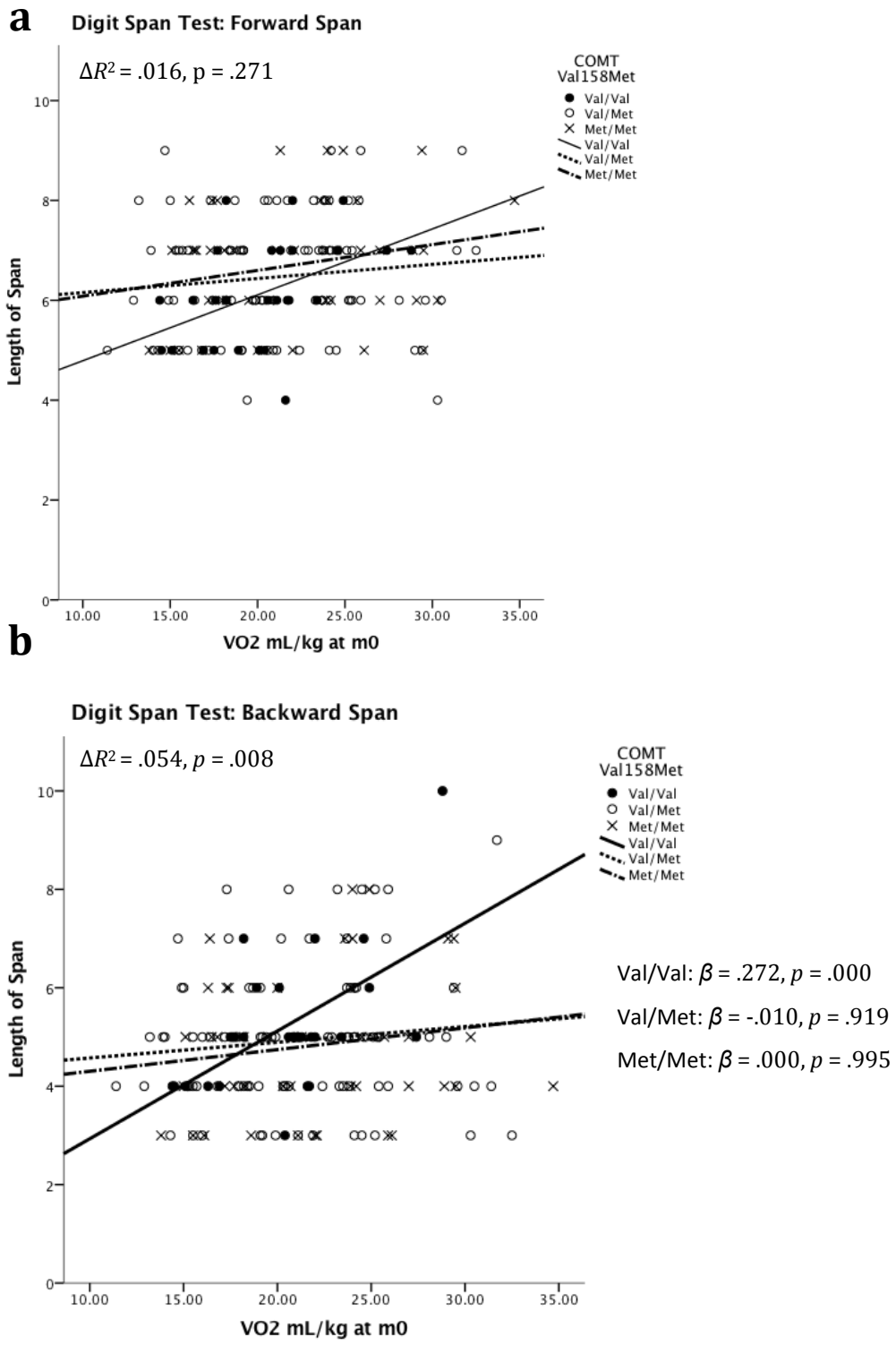


Figure 3.6 Interaction effect between COMT Val158Met and cardiorespiratory fitness level (i.e. VO_2) on length of span for the dual task test. **a)** Forward span: $\Delta R^2 = .016$, $\Delta F(2, 151) = 1.22$, p = .271 **b)** Backward span: $\Delta R^2 = .054$, $\Delta F(2, 151) = 4.99$, $p = .008$. Simple slopes: Val/Val, $B = 0.24$, $\beta = .272$, $t(151) = 3.20$, $p = .000$, 95% CI [0.09, 0.36]; Val/Met, $B = -0.00$, $\beta = -.010$, $t(151) = -0.12$, $p = .919$, 95% CI [-0.07, 0.08]; Met/Met, $B = 0.00$, $\beta = .000$, $t(151) = -0.01$, $p = .995$, 95% CI [-0.08, 0.09]

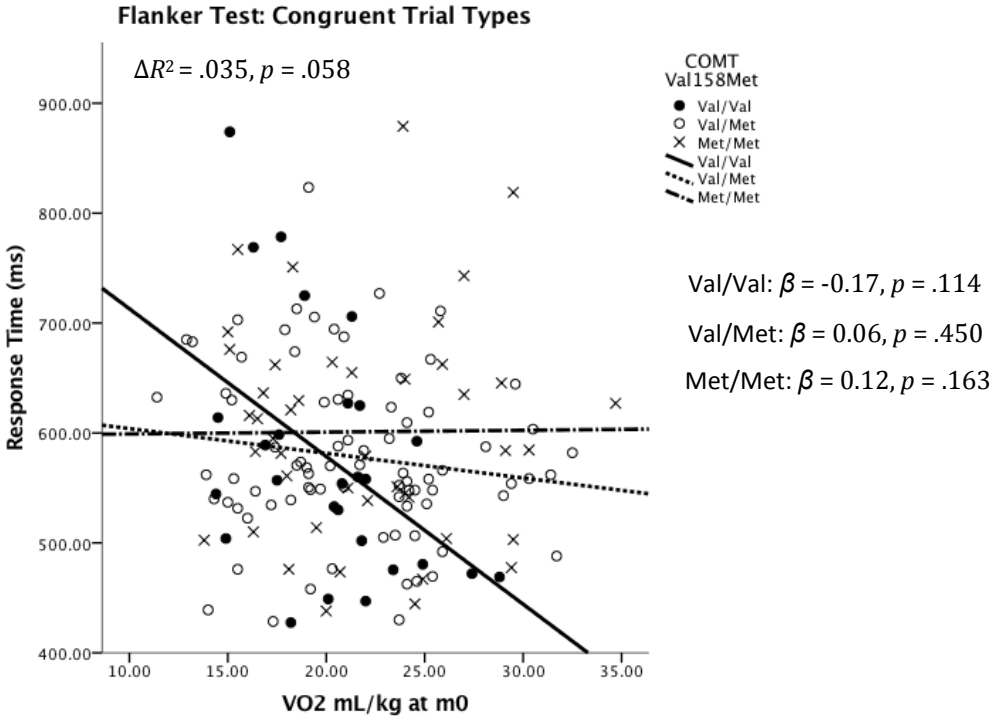


Figure 3.7 Interaction effect between COMT Val158Met and cardiorespiratory fitness level (i.e. VO_2) on response time for congruent trials on the Flanker test. The interaction marginally explained 3.5% of the variance in RT for congruent trials, $\Delta R^2 = .035$, $\Delta F(2, 143) = 2.90$, $p = .058$. Simple slopes were non-significant for all groups (see Table 3.18). However, the results suggest there was a difference in the direction of the relationship between response and cardiorespiratory fitness level between the groups: Val/Val vs Val/Met, $B = 10.24$, $\beta = .403$, $t(143) = 2.06$, $p = .060$, 95% CI [0.43, 22.87]; Val/Val vs Met/Met, $B = 12.61$, $\beta = .383$, $t(143) = 2.40$, $p = .031$, 95% CI [1.74, 25.63].

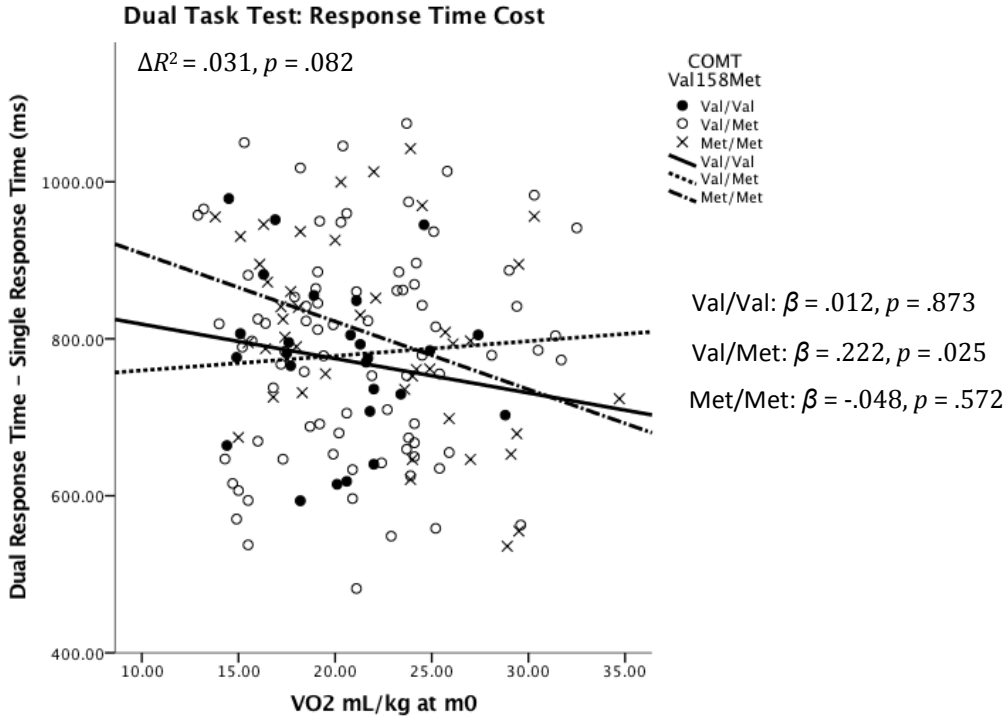


Figure 3.8 Interaction effect between COMT Val158Met and cardiorespiratory fitness level (i.e. VO_2) on response time cost for the dual task test. The interaction marginally explained 3.1% of variance in RT cost score, $\Delta R^2 = .031$, $\Delta F(2, 141) = 2.55$, $p = .082$. Simple slopes: Val/Val group, $B = 0.86$, $\beta = .012$, $t(141) = 0.13$, $p = .873$, 95% CI [-10.14, 11.17; Val/Met heterozygotes, $B = 7.99$, $\beta = .222$, $t(141) = 2.37$, $p = .025$, 95% CI [0.70, 14.75]; Met/Met, $B = -2.20$, $\beta = -.048$, $t(141) = -0.54$, $p = .572$, 95% CI [-9.40, 5.78].

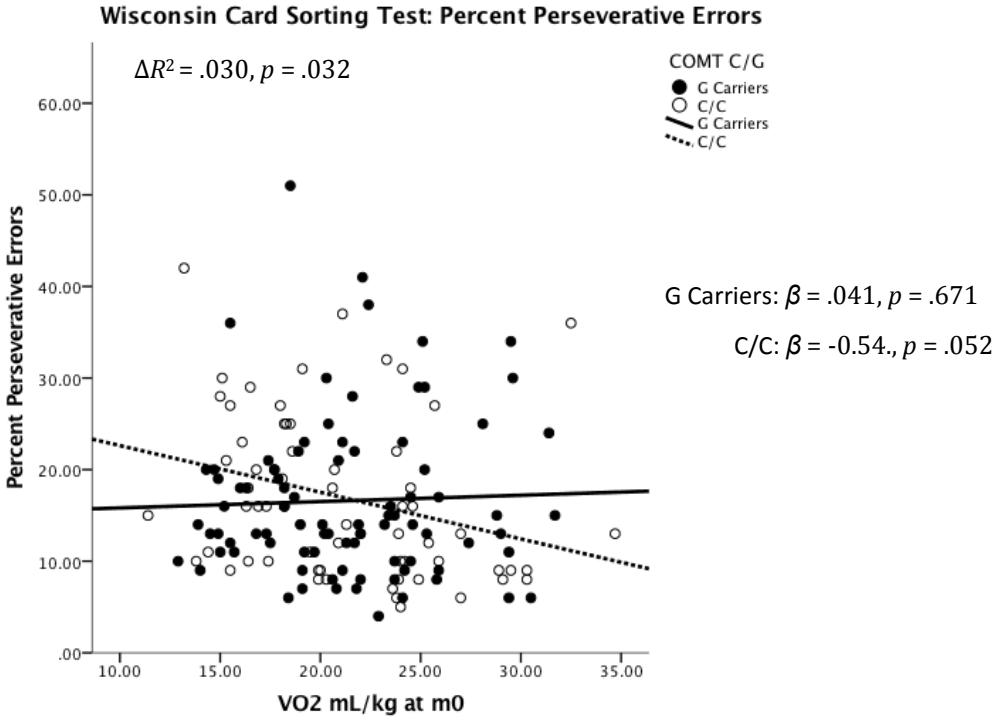


Figure 3.9 Interaction effect between COMT C/G and cardiorespiratory fitness level (i.e. VO_2) on performance measured by percent perseverative errors on the Wisconsin Card Sorting Test. The interaction significantly explained 3% of the variance in percent perseverative errors, $\Delta R^2 = .030$, $\Delta F(1, 142) = 4.68$, $p = .032$. Simple slopes: C/C group, $B = -0.54$, $\beta = -.203$, $t(142) = -2.22$, $p = .052$, 95% CI [-1.11, -0.29]; G Carriers, $B = 0.10$, $\beta = .041$, $t(142) = .430$, $p = .671$, 95% CI [-0.37, -0.59].

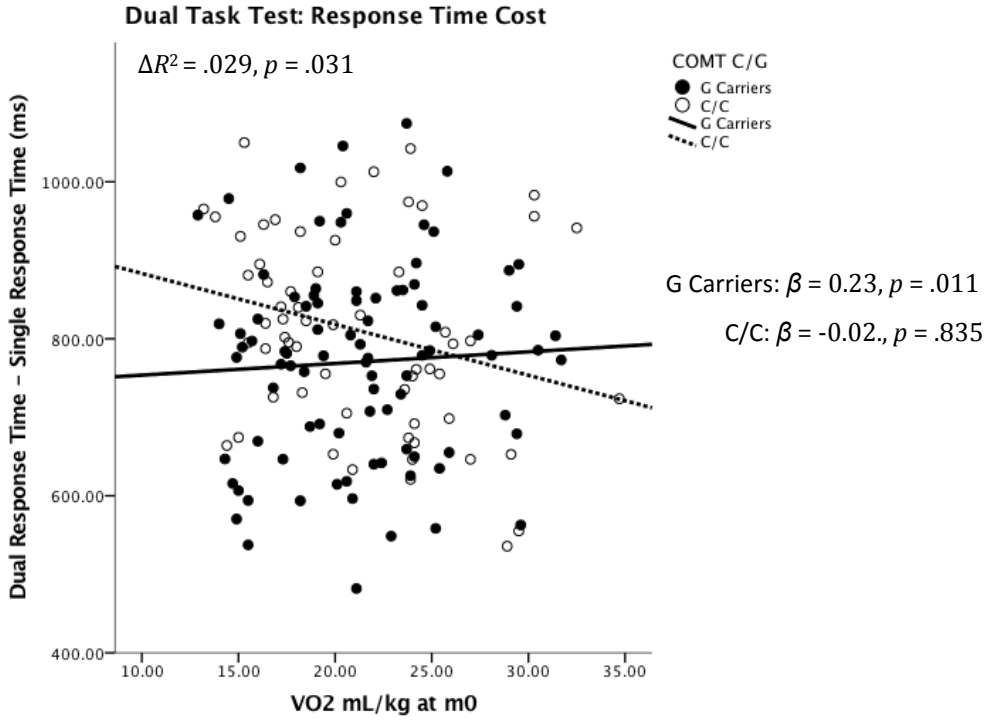


Figure 3.10 Interaction effect between COMT C/G and cardiorespiratory fitness level (i.e. VO_2) on a response time cost score for the dual task test. The interaction significantly explained 2.9% of the variance in a RT cost score, $\Delta R^2 = .029$, $\Delta F(1, 143) = 4.74$, $p = .031$. Simple slopes: G Carriers, $B = 8.30$, $\beta = .230$, $t(143) = 2.46$, $p = .011$, 95% CI [1.76, 14.59]; C/C Homozygotes, $B = -0.79$, $\beta = -0.02$, $t(143) = -0.23$, $p = .835$, 95% CI [-8.54, 6.31].

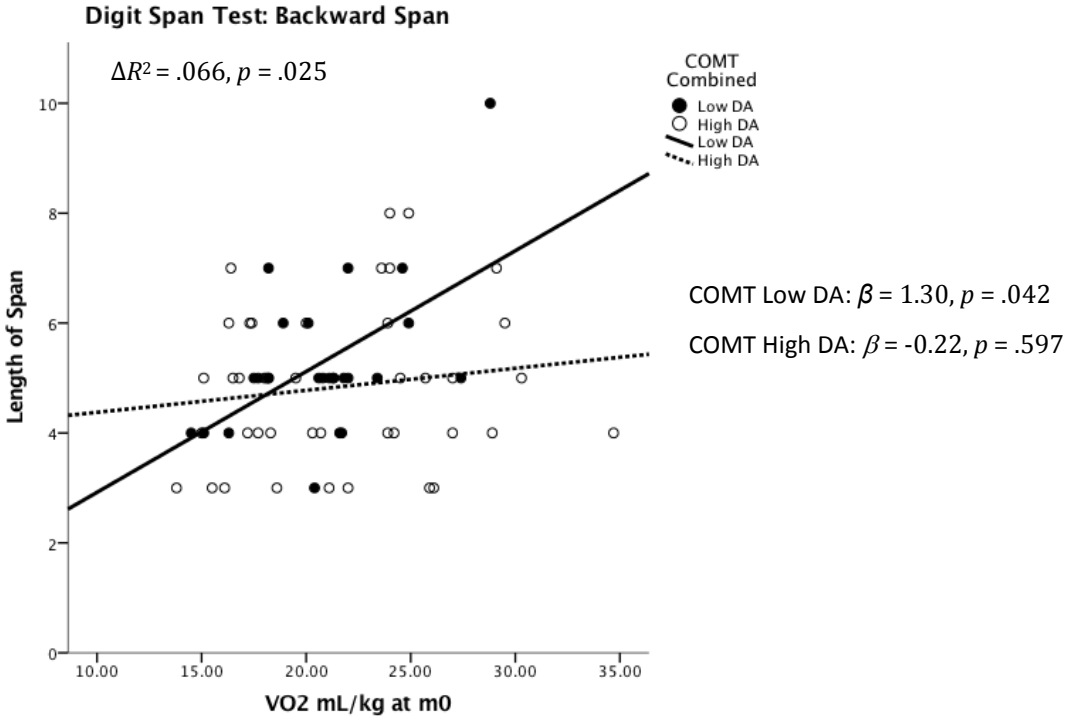


Figure 3.11 Interaction effect between COMT Combined and cardiorespiratory fitness level (i.e. VO_2) on the backward version of the digit span test. The interaction explained 6.6% of the variance in backward span, ΔR^2 = .066, $\Delta F(1, 60)$ = 5.31, p = .025. Simple slopes: Low DA group, B = 0.18, β = 1.30, $t(60)$ = 2.16, p = .042, 95% CI [-0.02, 0.33]; High DA group, B = -0.28, β = -0.22, $t(60)$ = -0.59, p = .597, 95% CI [-0.13, 0.08].

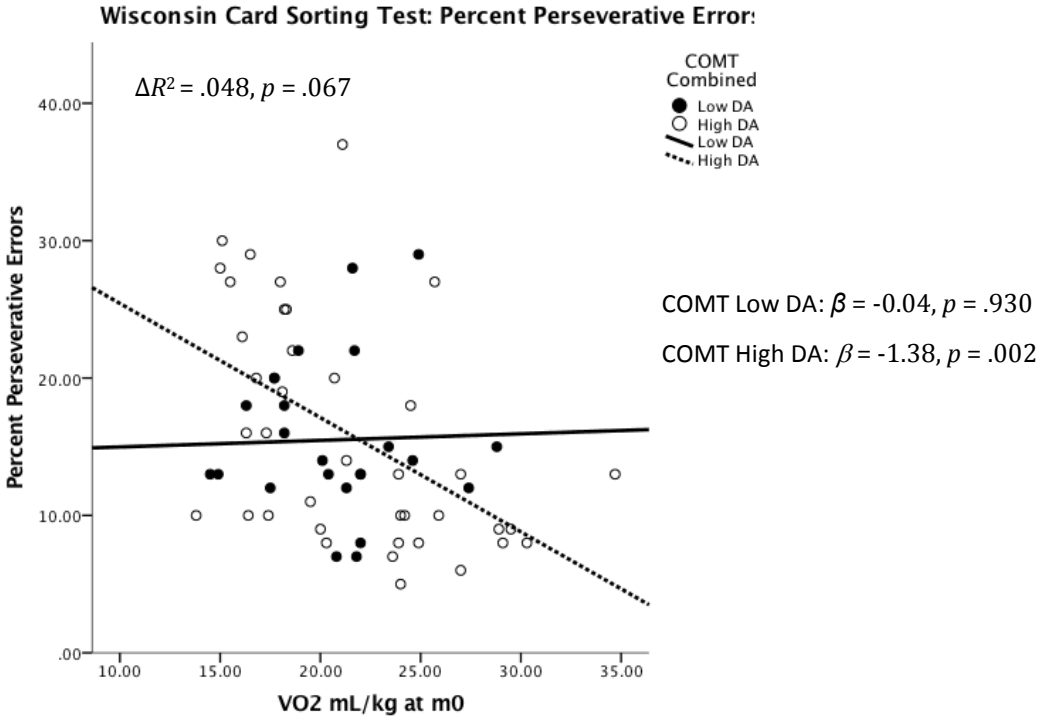


Figure 3.12 Interaction effect between COMT Combined and cardiorespiratory fitness level (i.e. VO_2) on performance of the Wisconsin Card Sorting Test measured by percent perseverative errors. The interaction marginally explained 4.8% of the variance in percent perseverative errors, $\Delta R^2 = .048$, $\Delta F(1, 56) = 3.49$, $p = .067$. Simple slopes: High DA group, $B = -0.92$, $\beta = -1.38$, $t(56) = -3.60$, $p = .002$, 95% CI [-1.54, -0.48]; Low DA group, $B = -0.32$, $\beta = -0.04$, $t(56) = -0.07$, $p = .930$, 95% CI [-0.69, 0.77].

Chapter 4: Growth Factor Genes

Growth factors are proteins that participate in the survival, growth and differentiation of developing neurons. These growth factors are widely distributed in the peripheral and central nervous systems, both during development and throughout the lifespan. Two growth factors of particular interest in this study are Brain-Derived Neurotrophic Factor (BDNF) and Insulin-like Growth Factor I (IGF1) because of their modulation by exercise and influence on cognition.

Methods

Sample

The initial sample size of *N* = 242 was reduced due to participant drop out, experimenter error, failure to complete behavioral or fitness testing, and genotyping failure. The sample size was then reduced further with pairwise deletion of participants that performed below chance (i.e. greater 50% error rate). The final sample size and group sizes are indicated within the results section. Due to the fact that there were only 2 Met/Met homozygotes, this group was combined with the Met/Val heterozygotes to conduct a Met carrier versus non-carrier analysis.

Genotyping

IGF1 G/A (rs6220). The IGF1 G/A polymorphism was assayed after nested PCR by using allele-specific T_m-shift primers in various combinations, together with automated melting curve analysis (Lipsky et al., 2001; Wang et al., 2005) on an iCycler real-time PCR machine (Bio-Rad, Hercules, CA). The yields of the PCR products corresponding to the A and G alleles were equalized by adjusting the concentrations of their respective oligonucleotide primers. Equalizing the yields of the two PCR products is necessary in part because they were designed to have different melting temperatures, and because accurate scoring requires detecting both alleles with equal sensitivity, particularly in heterozygotes. In this case, equalizing the yields also required the addition of DMSO (6.7%) and betaine monohydrate (333 mM) to the second round PCR buffer.

BDNF Val66Met (rs6265). The BDNF Val66Met polymorphism was also assayed after nested PCR by using allele-specific T_m-shift primers in various combinations, together with automated melting curve analysis (Lipsky et al., 2001; Wang et al., 2005) on an iCycler real-time PCR machine (Bio-Rad, Hercules, CA). The PCR product of the first round (outer) amplification was 184 bp in length. The PCR product of the second round (inner) amplification was 87 bp for the G allele, and 73 bp for the A allele.

Statistical Analysis

Hardy-Weinberg Equilibrium. Two hundred and forty samples were genotyped successfully for the IGF1 gene. The frequencies of the three IGF1 G/A genotypes were .10 for G/G, .38 for G/A, and .52 for A/A. The Hardy-Weinberg exact test showed that the allelic distribution did not deviate from expectations, $\chi^2(1) = 1.25$, $p = .275$.

Two hundred and forty-two samples were genotyped successfully for the BDNF gene. The frequencies of the three BDNF Val66Met genotypes were .90 for Val/Val (G/G), .09 for Val/Met (G/A), and .01 for Met/Met (A/A). The Hardy-Weinberg exact test showed the allelic distribution did not deviate from expectations, $\chi^2(1) = 3.16$, $p = .122$.

SNP analysis. All performance measures were analyzed using analysis of covariance (ANCOVA) with age, education, and sex as covariates. Percentile bootstrapped 95% confidence intervals (CI) were calculated over 5000 samples for pairwise comparisons and are reported in text if there was a significant omnibus effect of the SNP.

SNP x Fitness Level interaction analysis. All performance measures were analyzed using stepwise multiple regression with age, education, and sex entered in the first step as control variables. The SNP dummy variable(s) and cardiorespiratory fitness level (i.e. VO_2 scores) were both entered in the second step, and the SNP x Fitness Level interaction variable(s) was entered in the last step. If the SNP x Fitness Level interaction was significant (ΔR^2 was $p \leq .05$) or marginal ($p \leq .10$), simple slopes analysis was conducted to elucidate the nature of the interaction. For the simple slopes analysis, the control variables were entered in the first step, followed by the SNP dummy variable(s) in the second step, and then in the last step cardiorespiratory fitness level was added for each individual group (e.g. VO_2 scores for Met Carriers with Val/Val Homozygotes coded 0 and

VO_2 scores for Val/Val Homozygotes with Met Carriers coded 0 as two separate interaction variables for the BDNF Val66Met SNP).

Cognitive Assessment

This section is included here to help remind the reader of the variables of interest for each particular test. Full descriptions of the tests can be found in Chapter 2: General Methods.

Verbal crystallized intelligence. One test was used to asses verbal crystallized intelligence.

K-BIT test. The primary measure was an age-scaled score of correct items divided by the total number of items attempted on the verbal subscale of the Kaufman Brief Intelligence Test (K-BIT; Kaufman & Kaufman, 1990). Higher scores indicate better performance.

Memory. Two tests were used to assess various aspects of memory (i.e. short-term, working, and spatial).

Digit span test. Two versions of the digit span test, forward and backward, were used to assess short-term and working memory, respectively. For both versions of the digit span, the primary measure was the total number of digits in the maximum span length repeated; therefore, higher scores indicate better performance.

Spatial memory test. The primary measures were response time (RT) and error rate (ER) for each of the three memory loads (i.e. 1, 2 or 3). Lower scores (i.e. faster RT and lower ER) indicate better performance.

Executive control functioning. Six tests were used to assess various aspects of executive control functioning.

Verbal fluency test (FAS). The verbal fluency test assessed fluid intelligence and the primary measure was the total number of words named in 180 seconds for three phonetic categories (i.e. 60 seconds for F, A, & S separately). Higher scores indicate better performance.

Flanker test. The Flanker test assessed inhibitory processing. The primary measures were RT and ER for the two trial types (i.e. congruent and incongruent).

Comparison of these two trial types assessed the ability to inhibit conflicting information. Lower scores indicate better performance.

Wisconsin card sorting test. The Wisconsin Card Sorting Test (WCST) assessed many aspects of executive functioning including inhibitory processing and concept shifting. The primary measures were percent perseverative errors and total errors; therefore, lower scores indicate better performance.

Task switch test. The task switch test assessed maintenance and coordination of multiple sets. The primary measures were RT and ER for the two conditions (i.e. single and mix), which can be compared to assess the cost of maintaining one versus two rule sets. The mix condition can be further split into repeat and switch trial types, which can be compared to assess the cost of rule switching. Lower scores indicate better performance.

Dual task test. The dual task test assessed coordination of multiple sets. The primary measures were RT and ER for the two trial types (i.e. single and dual), which can be compared to assess the cost of responding to one versus two stimuli. Lower scores indicate better performance.

Insulin-Like Growth Factor

Insulin-like growth factor I (IGF1) belongs to a family of growth factors known as somatomedins that play an important role in mammalian growth and development (Froesch, Schmid, Schwander, & Zapf, 1985). Although all tissues express IGFs (Fernandez & Torres-Alemán, 2012), 90% of the body's total IGF1 is synthesized and steadily secreted by the liver into the serum where it is bound by specific carrier proteins, insulin-like growth factor binding proteins (IGFBP), allowing it to be present in high concentrations within the serum without causing hypoglycemia (Froesch et al., 1985). It is generally considered to be a peripheral peptide hormone that regulates fuel metabolism (Fernandez & Torres-Alemán, 2012), but also exerts neurotrophic and neuroprotective effects in the central nervous system (CNS) by stimulating the viability and functioning of different neuronal and glial cell types (van Dam et al., 2000; van Dam & Aleman, 2004).

Serum IGF1 is able to cross the blood brain barrier (Reinhardt & Bondy, 1994) into active brain regions through brain vessels by means of an activity-dependent mechanism

that is coupled to neuronal activity (Nishijima et al., 2010). This crossing is mediated by IGF1 receptors that are highly expressed in endothelial cells of blood vessels in the brain (Bondy & Lee, 1993) allowing for transcytosis of IGF1 (i.e. regulated endocytosis and transport from one side of the cell to the other for eventual release; Fernandez & Torres-Alemán, 2012). IGF1 can also enter the cerebrospinal fluid (CSF) flow through the same mechanism since IGF1 receptors are also highly expressed in epithelial cells of the choroid plexus (Bondy & Lee, 1993). These two methods of entry from the circulation into the brain suggest that peripheral IGF1 may mediate actions of IGF1 in various brain areas. Not surprisingly, an increase in IGF1 blood levels in the periphery leads to increased levels in the CSF (Carro, Nez, Busiguina, & Torres-Aleman, 2000), and once in the CSF, IGF1 can easily diffuse to periventricular brain areas, such as the hippocampus. However, IGF1 does not only operate through an endocrine system (Fernandez & Torres-Alemán, 2012). Local synthesis also occurs within all cell types of the brain (Fernandez & Torres-Alemán, 2012; Han, 1995), and the IGF1 receptor has been detected in neuronal nuclei where it is thought to affect gene transcription directly by acting as a transcriptional modulator (Sehat et al., 2010).

Indicative of its importance in brain development and overall brain health, IGF1 has a diffuse expression pattern including the cortex, hippocampus, cerebellum, brainstem, hypothalamus and the spinal cord (Bach, Shen-Orr, Lowe, Roberts, & Leroith, 1991). Experimentally, IGF1 has been shown to rescue degenerating motoneurons after axotomy in young mice (Li, Oppenheim, Lei, & Houenou, 1994), and increased IGF1 levels are also observed in experimental animals during recovery from brain injuries caused by trauma (Garcia-Estrada, Garcia-Segura, & Torres-Aleman, 1992), hypoxic-ischaemic injury (Guan, Williams, Skinner, Mallard, & Gluckman, 1996; Guan, Williams, Gunning, Mallard, & Gluckman, 1993), or induced by electrolytic lesions (Yamaguchi et al., 1991). On the other hand, knocking out IGF1 in mice impairs neuronal somatic and dendritic growth (Cheng et al., 2003).

In concert with IGF1's support of overall brain health, there is evidence for a similar support of cognitive ability. For example, in liver-IGF-1 deficient mice, IGF1 administration can restore adult hippocampal neurogenesis and improve spatial memory (Trejo, Llorens-Martín, & Torres-Alemán, 2008). Further, human patients with growth hormone

deficiencies show reduced cognitive performance, in part due to severe reductions in IFG1 levels in both plasma and the CNS (van Dam et al., 2000). Experimentally, several studies of healthy adults have found positive associations between IGF1 and cognitive functions, including working memory (Bellar, Glickman, Juvancic-Heltzel, & Gunstad, 2011), as evidenced in a meta-analysis conducted by (Arwert, Deijen, & Drent, 2005).

Developmentally, high concentrations of IGF1 are observed during puberty, with a sharp decline during the first years after puberty and further reduction between the ages of 30 to 75 (van Dam & Aleman, 2004). This age-related decline, along with its role in cognition, implicate a potential role for IGF1 in aging-related cognitive decline (Aleman et al., 1999; Markowska, Mooney, & Sonntag, 1998; van Dam & Aleman, 2004). In line with this, Morely et al. (1997) found significant correlations between IGF1:GH ratio and cognitive performance, including visual and verbal memory in a sample of 56 men aged 20 to 84. In older populations, lower levels of IGF1 are associated with lower levels of cognitive functions such as processing speed, memory, and executive functioning (for review see van Dam & Aleman, 2004). For example, Aleman et al. (1999) found a positive correlation between circulating IGF1 levels and speed of information processing, measured by performance on a digit symbol substitution and a concept-shifting task (similar to Trails B) in a sample of 25 men between 65 to76 years of age. Vitiello et al. (2006) found a positive association between IGF1 levels and age-sensitive problem-solving abilities in elderly men and women (aged 60 to 85 years).

Furthermore, Dik et al. (2003) found a cross sectional assocation between IGF1 levels and information processing speed and moreover, low IGF1 levels were associated with a 3-year decline in information processing speed in older adults aged 65 to 88 years. This evidence suggests IGF1 levels may have a protective effect against aging related cognitive decline (Aleman & Torres-Alemán, 2009). In line with this, research in non-human animals has shown that intraventricular infusion of IGF1 can improve working memory in a repeated acquisition task, an object recognition task, and a place discrimination task in aged rats, but does not affect sensorimotor skills or exploration (Markowska et al., 1998).

Although an age-related decline in circulating levels of IGF1 is a general trend, there are substantial individual differences in existing IGF1 levels (van Dam & Aleman, 2004). In

fact, 40-60% of variation in circulating IGF1 levels is determined by genetic factors (Hall, Hilding, & Thorén, 1999; Harrela et al., 1996; Hong, Pedersen, Brismar, Hall, & de Faire, 1996; Verhaeghe et al., 1996). The *IGF1* gene is located on chromosome 12q and there is a common SNP resulting in a G to A switch in exon 4 of the 3' untranslated region (rs6220; http://www.ncbi.nlm.nih.gov/SNP/snp_ref.cgi?rs=rs6220). This region contains regulatory domains for mRNA expression and may alter the RNA stability and regulation of protein expression (Al-Zahrani et al., 2006; Huuskonen et al., 2011). This SNP has been associated with circulating levels of IGF1 protein such that the minor G allele is associated with higher levels of circulating IGF1 (Diorio, Brisson, Bérubé, & Pollak, 2008; Johansson et al., 2007; Verheus et al., 2008). Therefore, this study was conducted to investigate whether the IGF1 G/A SNP would have an effect on cognitive performance in healthy elderly adults due to its influence on circulating levels of IGF1.

A low expression of IGF1 mRNA, but a wide expression of IGF1 receptors in the adult mammalian brain, suggests that peripherally produced IGF1 plays an important role in the adult brain (Carro et al., 2000; Trejo, Carro, & Torres-Aleman, 2001). Generally, IGF1 levels in the serum are primarily regulated by growth hormone (Froesch et al., 1985), but levels can also be moderated by other factors such as exercise. Exercise has been shown to increase the expression of IGF1, BDNF, enhance learning acquisition, and augment recall in rats (Cotman, Berchtold, & Christie, 2007). Interestingly, blockade of the IGF1 receptor diminishes exercise-induced increases in levels of BDNF and eliminates the augmentation of recall , indicating that IGF1 may modulate exercise-induced benefits (Carro et al., 2000; Carro, Trejo, Busiguina, & Torres-Aleman, 2001; Trejo et al., 2001, 2008). Therefore, we also investigated the possibility of an interaction between IGF1 G/A and cardiorespiratory fitness levels in the same subjects.

IGF1 G/A (rs6220) Predictions

This study takes two novel approaches to investigate the relationship between IGF1 and cognition. It will be the first time to date that the IGF1 gene and, furthermore, the interaction between the IGF1 gene and cardiorespiratory fitness level has been related to cognition. For the IGF1 G/A SNP, since the A allele is associated with lower levels of

circulating IGF1, I predict A carriers will show poorer cognitive performance on tests of memory and executive control, but show a positive relationship between cardiorespiratory fitness level and behavioral performance. This prediction is extrapolated from the findings in the extant literature showing: 1) an association between the IGF1 gene and IGF1 levels; 2) evidence that IGF1 levels can be modulated by exercise; and 3) a positive association between circulating IGF1 levels and cognition.

IGF1 G/A (rs6220) Results

There were no significant differences between the genotypic groups on age, education, mMMSE, BMI or VO_2 (Table 4.1). Chi-square analysis showed the distribution of males and females was not significantly different between the groups (Table 4.1). Task means and standard deviations are reported in Table 4.3.

IGF1: Verbal Crystallized Intelligence

Verbal crystallized intelligence has been shown to remain relatively stable in older adult populations (Hedden & Gabrieli, 2004; Park & Gutchess, 2002). Due to the relative stability of the verbal crystallized intelligence, I do not expect to find an effect of IGF1 G/A or its interaction with cardiorespiratory fitness level.

KBIT test. The sample size for the KBIT test was N = 192, with genotypic group sizes as follows: A/A = 101, G/A = 71, G/G = 20. As expected, there was no significant difference between the IGF1 G/A genotypic groups on the age-scaled scores $F(2, 186) < 1$, suggesting that IGF1 G/A genotype does not influence verbal crystallized intelligence.

When cardiorespiratory fitness level was added to the model, the sample size for the KBIT test was reduced to N = 158, with genotypic group sizes as follows: A/A = 79, G/A = 60, G/G = 19. Hierarchical regression showed the interaction between IGF1 G/A x Fitness Level did not explain a significant proportion of variance in the age-scaled score, ΔR^2 = .012, $\Delta F(2, 149) = 1.10$, p = .336. This was expected since neither IGF1 nor cardiorespiratory fitness level were predicted to affect verbal crystallized intelligence.

IGF1: Memory Tests

Performance on tests of working and spatial memory are seen to decrease in older adult populations, in contrast to more stable performance on tests of short-term memory (Hedden & Gabrieli, 2004; Park & Gutchess, 2002). In terms of the IGF1 G/A SNP, the G allele has been associated with higher levels of circulating IGF1 (Diorio et al., 2008; Johansson et al., 2007; Verheus et al., 2008), and in general, there is a positive association between IGF1 levels and cognitive functioning (Arwert et al., 2005; Bellar et al., 2011). Therefore, I predict G carriers will perform better on the various memory tests. Further, the extant literature shows that exercised-induced increases in both memory performance and IGF1 levels, and that blocking IGF1 receptors can block exercise-induced behvavioral gains. Thus, I expect to find an interaction between the IGF1 G/A SNP and cardiorespiratory fitness levels such that A allele carriers will show the strongest relationship between cardiorespiratory fitness level and memory performance.

Digit span test. The sample size for the digit span test was $N = 194$, with genotypic group sizes as follows: A/A = 102, G/A = 72, G/G = 20. No significant differences were found between the IGF1 genotypic groups for either forward span, $F(2, 188) = 1.44$, $p = .204$, or backward span, $F(2, 188) < 1$. This was expected for the forward version of the digit span, a test of short-term memory, but I expected the A/A group would perform better on the backward version, a test of working memory.

When cardiorespiratory fitness level was added into the model the sample size for the digit span tests was reduced to $N = 160$, with genotypic group sizes as follows: A/A = 80, G/A = 61, G/G = 19. Hierarchical regression showed no interaction effect of IGF1 G/A x Fitness Level on either forward span, $\Delta R^2 = .009$, $\Delta F(2, 151) < 1$, or backward span, $\Delta R^2 = .024$, $\Delta F(2, 151) = 2.09$, $p = .128$. Again, this was expected for forward span, but not for backward span. Based on previous studies that showed increasing fitness levels can increase IGF1 levels, and that the IGF1 G/A genotype influences IGF1 levels, I expected a potential interaction between the two variables.

Spatial memory test. The sample size for the spatial memory test was $N = 175$, with genotypic group sizes as follows: A/A = 94, G/A = 67, G/G = 14. The G/A group showed the fastest response times (RT) across all memory loads, while the G/G group (expected to perform the best) were slowest (Figure B.1). This difference was significant

for memory load 1, $F(2, 169) = 3.46$, $p = .034$, and memory load 3, $F(2, 169) = 4.30$, $p = .015$. Although the differences between genotypic groups were not significant for memory load 2, $F(2, 169) = 1.35$, $p = .262$, the groups followed the same trend with the G/A group showing the fastest response times and the G/G group the slowest. Bootstrapped pairwise comparisons are reported in Table B.1. The genotypic groups did not differ on error rate (ER) for any Memory Load, all $F(2, 169) < 1$, suggesting differences in RT were not due to an accuracy versus speed trade off. These results suggest that the IGF1 G/A genotype influences spatial memory (although not in the expected direction), but not short-term or working memory.

Addition of cardiorespiratory fitness level reduced the sample size for the spatial memory test to $N = 144$ with genotypic group sizes as follows: A/A = 75, G/A = 56, G/G = 13. The interaction between IGF1 G/A x Fitness Level significantly explained 4.7% of the variation in error rate for memory load 2, $\Delta R^2 = .047$, $\Delta F(2, 135) = 3.56$, $p = .031$. Simple slopes analysis revealed that increasing VO_2 led to decreasing errors only for the G/A group, $B = -.008$, $\beta = -0.88$, $t(135) = -2.40$, $p = .037$, 95% CI [-.015, .000], but there was a non-significant relationship for both the A/A, $B = -.001$, $\beta = -0.10$, $t(135) = -0.33$, $p = .682$, 95% CI [-.005, .003], and G/G group, $B = .008$, $\beta = 0.53$, $t(135) = 1.37$, $p = .181$, 95% CI [-.007, .022] (Figure 4.1). The interaction did not significantly explain variance in error rate or response time for any other memory loads (Table 4.3).

IGF1: Executive Control Function Tests

Increasing age most negatively affects performances on tests requiring some form of executive control (Hedden & Gabrieli, 2004; Park & Gutchess, 2002), but higher levels of fitness tend to be associated with better executive control (Colcombe & Kramer, 2003). As mentioned in the previous section, since the G allele of the IGF1 G/A SNP is been associated with higher levels of circulating IGF1 (Diorio et al., 2008; Johansson et al., 2007; Verheus et al., 2008), and there is a positive association between IGF1 levels and cognitive functioning (Arwert et al., 2005; Bellar et al., 2011), I again predict G carriers will perform better on the various tests of executive control. I also expect to find an interaction between the IGF1 G/A

SNP and cardiorespiratory fitness levels such that A allele carriers will show the strongest relationship between cardiorespiratory fitness level and executive control performance.

Stroop test. The sample size for the Stroop test was N = 157 with genotypic group sizes as follows: A/A = 80, G/A = 59, G/G = 18. ANCOVA showed a significant difference between the groups on RT for the incongruent-ineligible trial types, $F(2, 151) = 3.08$, p = .049 (Figure B.2). Bootstrapped pairwise comparisons showed the A/A group was significantly slower than the G/G group, mean difference = 57.71, 95% CI [14.95, 100.35], and marginally slower than the G/A group, mean difference = 29.74, 95% CI [-5.21, 63.73]. The groups did not significantly differ on RT for any other trial type but did follow the same trend with the A/A group showing the slowest RTs, neutral RT, $F(2, 151) = 2.24$, p = .110; congruent RT, $F(2, 151) < 1$; incongruent eligible RT, $F(2, 151) = 1.60$, p = .204. There were also no significant differences between the genotypic groups on error rate (ER) for any of the trial types: neutral ER, $F(2, 151) < 1$; congruent ER, $F(2, 151) = 1.14$, p = .332; incongruent-ineligible ER, $F(2, 151) = 2.17$, p = .118; incongruent eligible ER, $F(2, 151) = 1.56$, p = .213. This follows predictions that the G allele carriers will show greater cognitive performance.

When cardiorespiratory fitness level was added to the model the sample size for the Stroop test was reduced to N = 151, with genotypic group sizes as follows: A/A = 75, G/A = 58, G/G = 18. Hierarchical regression showed the interaction between IGF1 G/A x Fitness Level significantly explained variance in response time for each trial type (Table 4.4): neutral RT, $\Delta R^2 = .066$, $\Delta F(2, 142) = 5.50$, p = .005 (Figure 4.2a); congruent RT, $\Delta R^2 = .041$, $\Delta F(2, 142) = 3.29$, p = .040 (Figure 4.2b); incongruent-ineligible RT, $\Delta R^2 = .040$, $\Delta F(2, 142) = 3.28$, p = .041 (Figure 4.2c); incongruent eligible RT, $\Delta R^2 = .038$, $\Delta F(2, 142) = 3.08$, p = .049 (Figure 4.2d). Simple slopes analysis showed RT significantly decreased with increasing VO_2 only for the G/A group for each trial type, neutral RT, B = -9.15, β = -1.14, $t(142) = -3.16$, p = .002, 95% CI [-15.31, -3.58], congruent RT, B = -7.01, β = -0.91, $t(142) = -2.48$, p = .028, 95% CI [-13.83, -1.32], incongruent-ineligible RT, B = -8.31, β = -0.88, $t(142) = -2.42$, p = .012, 95% CI [-14.85, -1.42], incongruent eligible RT, B = -8.98, β = -0.88, $t(142) = -2.40$, p = .016, 95% CI [-16.89, -2.01] (Figure 4.2; other simple slopes reported in Table 4.5). There was also a marginal interaction effect on error rate for the incongruent eligible trial type,

ΔR^2 = .033, $\Delta F(2, 142) = 2.83$, p = .062 (Figure 4.3d). Simple slopes analysis again revealed that error rate decreased with increasing VO_2 only for the G/A group, B = -.006, $t(142)$ = -1.49, p = .052, 95% CI [-.013, -.000], but a non-significant relationship in the A/A group. The interaction did not significantly explain variance for any other trial type, but the trend was similar (Figure 4.3a-c) These results suggest that the moderate level of IGF1 benefits the most from increasing VO_2.

Task switch test. The sample size for the task switch test was N = 157 with genotypic group sizes as follows: A/A = 84, G/A = 55, G/G =18. ANCOVA showed no significant differences between genotypic groups on response time for either condition, single or mix, both $F(2, 151) < 1$. There were also no differences between the groups on a global RT cost (i.e. mix RT – single RT), $F(2, 151) < 1$. There was, however, a marginal difference on error rate (ER) in the single condition, $F(2, 151) = 2.72$, p = .069, but bootstrapped pairwise comparisons showed no significant differences between the groups (Table B.2; Figure B.3a). There were no differences in ER in the mix Condition, $F(2, 151)$ = 2.14, p = .121.

Further analysis of the Mix Condition showed no differences between the groups on RT for either trial type: repeat, $F(2, 151) < 1$; switch, $F(2, 151) = 1.04$, p = .357. There were also no group differences on a local RT cost (i.e. switch RT – repeat RT), $F(2, 151)$ =2.13, p = .123. Although there were no differences on error rate (ER) for repeat trials, $F(2, 151)$ = 1.44, p = .241, there was a marginal difference on ER for switch trials, $F(2, 151) = 2.64$, p = .074 (Figure B.3b). Bootstrapped pairwise comparisons revealed the G/G group had significantly more errors compared to the G/A group, mean difference = .064, 95% CI [.003, .125], and the A/A group, mean difference = .073, 95% CI [.009, .137]. Although there were no significant differences between the groups on RT, the G/G group showed the highest RT and a significantly higher error rate, so there was no accuracy versus speed tradeoff. This suggests that the G/G group had a more difficult time switching between two task sets.

When cardiorespiratory fitness level was added into the model, the sample size was reduced to N = 131 with group sizes as follows: A/A = 68, G/A = 45, G/G = 18. The interaction between IGF1 G/A x Fitness Level did not significantly explain RT in either the single, ΔR^2 = .047, $\Delta F(2, 122) = 3.08$, p = .050 (the overall model was not significant, $F(8, 122) = 1.04$, p = .409), or mix Condition, ΔR^2 = .012, $\Delta F(2, 122) < 1$. The interaction did

marginally explain 3.6% of the variance in ER for the single condition, ΔR^2 = .036, ΔF(2, 122) = 2.54, p = .083 (Figure 4.4), but not the mix condition, ΔR^2 = .005, ΔF(2, 122) < 1. Simple slopes analysis revealed ER significantly decreased with increasing VO_2 for the G/G group, B = -.012, β = -0.99, t(122) = -2.94, p = .049, 95% CI [-.023, .000], but there was no significant relationship for either the G/A, B = -.005, β = -0.63, t(122) = -1.61, p = .156, 95% CI [-.012, .003], or the A/A group, B = -.002, β = -0.29, t(122) = -0.87, p = .350, 95% CI [-.007, .002].

Further analysis of the mix condition showed the interaction marginally explained 4.3% of the variance in RT for repeat trials, ΔR^2 = .043, ΔF(2, 122) = 3.02, p = .052 (Figure 4.5), but not for switch trials, ΔR^2 = .003, ΔF(2, 122) < 1 (Figure 4.5). Simple slopes analysis showed RT significantly decreased with increasing VO_2 for the G/A group, B = -11.31, β = -.607, t(122) = -1.70, p = .041, 95% CI [-22.95, -1.07], but not for the A/A, B = 2.65, β = .149, t(122) = 0.49, p = .673, 95% CI [-10.19, 14.39], or the G/G group, B = -8.49, β = -.288, t(122) = -0.92, p = .199, 95% CI [-26.64, 9.66]. There was no interactive effect on ER for either repeat, ΔR^2 = .007, ΔF(2, 122) < 1, or switch trials, ΔR^2 = .004, ΔF(2, 122) < 1. There was also no effect on local cost scores for RT, ΔR^2 = .028, ΔF(2, 122) = 1.92, p = .151, or ER, ΔR^2 = .006, ΔF(2, 122) < 1.

Dual task test. The sample size for the dual task test was N = 156, with genotypic group sizes as follows: A/A = 79, G/A = 59, G/G = 18. ANCOVA showed no significant differences between the genotypic groups on response time (RT) for either the single, F(2, 150) < 1, or dual trial types, F(2, 150) = 1.69, p = .189 (Figure B.4a). However, there was a significant difference between the groups on a RT cost score (i.e. dual RT – single RT), F(2, 150) = 3.87, p = .023 (Figure B.4b). Bootstrapped pairwise comparisons revealed the A/A group showed a significantly higher cost than the G/G group, mean difference = 82.95, p = .016, 95% CI [15.70, 152.60], and marginally higher cost compared to the G/A group, mean difference = 36.33, p = .078, 95% CI [-3.23, 16.12]. There was also a significant difference between the genotypic groups on error rate for single trials, F(2, 150) = 3.05, p = .050, and a marginal difference on dual trials, F(2, 150) = 2.42, p = .092 (Figure 4.8c). Bootstrapped pairwise comparisons for single trials revealed the G/A group showed significantly higher error rates compared to the G/G group, mean difference = .023, p = .069, 95% CI [-.002,

.048], and marginally higher than the A/A group, mean difference = .023, p = .069, 95% CI [-.002, .048]. For dual trials, bootstrapped pairwise comparisons revealed the G/G group had significantly lower error rates compared to both the A/A group, mean difference = -.062, p = .003, 95% CI [-.100, -.026], and the G/A group, mean difference = -.078, p = .004, 95% CI [-.125, -.029]. This suggests the A/A group was having more difficulty with the dual task, in maintaining the rules and coordinating two responses.

When cardiorespiratory fitness level was added into the model, the sample size was reduced to N = 150 with group sizes as follows: A/A = 75, G/A = 58, and G/G = 17. Hierarchical regression showed the interaction between IGF1 G/A x Fitness Level did not significantly explain variance in RT for single trials, ΔR^2 = .023, $\Delta F(2, 141) = 1.75$, p = .178, but marginally explained 2.8% of the variance in RT for dual trials, ΔR^2 = .028, $\Delta F(2, 141) = 2.35$, p = .099. Simple slopes analysis revealed that dual RT marginally decreased with increasing VO_2 for the G/G group, B = -14.26, β = -0.48, $t(141) = -1.52$, p = .058, 95% CI [-30.98, 0.27] (Figure 4.6). The relationship was not significant for the G/A group, B = -8.48, β = -0.46, $t(141) = -1.33$, p = .154, 95% CI [-21.14, 2.62] or the A/A group, B = 3.60, β = 0.21, $t(141) = 0.70$, p = .559, 95% CI [-8.50, 15.60]. Also, the interaction did not explain variance in error rate (ER) for either single, ΔR^2 = .001, $\Delta F(2, 141) < 1$, or dual trials, ΔR^2 = .007, $\Delta F(2, 141) < 1$.

Brain-Derived Neurotrophic Factor

Brain-derived neurotrophic factor (BDNF) belongs to a family of growth factors called neurotrophins. Neurotrophins are regulatory factors that mediate the differentiation, proliferation and survival of cholinergic, dopaminergic and serotonergic neurons (Savitz et al., 2006). BDNF exerts long-term effects on neuronal survival, migration, and dendritic and axonal growth mostly in the prefrontal cortex (PFC) and hippocampus (HC), even though it is expressed throughout the brain (Savitz et al., 2006). BDNF also displays neurotrophic and neuroprotective properties and can affect functions that underlie brain plasticity (Meeusen, 2005). For example, it is known to be important in modulating synaptic changes, such as hippocampal long-term potentiation (LTP), which is associated with learning and memory (Egan et al., 2003; Hariri et al., 2003). For these

reasons, BDNF is considered a major contributor to alterations in hippocampal function and hippocampal-dependent learning and memory and has also been implicated in neurogenesis in the HC (Egan et al., 2003; Hariri et al., 2003; Savitz et al., 2006). In rodents, blocking either the release of BDNF or the binding of BDNF to its receptor TrkB eliminates LTP in the HC (for review see Pang & Lu, 2004). BDNF specifically moderates synaptic plasticity and neurogenesis in the dentate gyrus and has been directly related to learning rates in spatial memory paradigms (Hwang et al., 2006; Silhol, Arancibia, Maurice, & Tapia-Arancibia, 2007).

A frequent functional SNP in the BDNF gene that swaps a G to an A at nucleotide 196, results in a valine (Val) to methionine (Met) amino acid substitution at codon 66 (Val66Met; rs6265). Although this SNP is located in the 5' pro-BDNF sequence, making it unlikely to affect the actual biological activity of the mature protein, it is believed to alter the intracellular processing and regulated activity-dependent secretion of mature BDNF (Egan et al., 2003). The Met allele is thought to selectively impair secretion and intracellular trafficking of BDNF in primary cortical neurons and neurosecretory cells (Egan et al., 2003; Savitz et al., 2006).

To further the understanding of how Val66Met may elicit *in vivo* phenotypes, Egan et al. (2003) examined Val- and Met-BDNF expression and secretion using cultured hippocampal neurons. They found that Val-BDNF was mostly localized in cell bodies and dendrites, whereas Met-BDNF was primarily localized in cell bodies and only in a few proximal dendrites. They also found that while constitutive secretion of Met-BDNF and Val-BDNF did not differ, secretion of Met-BDNF after neuronal depolarization was severely attenuated. Properties and biological function of the secreted Val- and Met-BDNF did not differ and both were successfully targeted into Golgi, but only Val-BDNF seems to be sorted from Golgi to secretory granules and localized to synapses.

Several studies have also shown that diminished secretion in Met carriers is related to reductions in hippocampal and prefrontal grey matter volume (Bueller et al., 2006; Erickson et al., 2010; Pezawas et al., 2004), alterations in hippocampal function (Egan et al., 2003; Hariri et al., 2003), and cognitive impairments (Egan et al., 2003; Hariri et al., 2003; Miyajima et al., 2008; Rybakowski, Borkowska, Czerski, Skibińska, & Hauser, 2003). Further, BDNF levels decline with advancing age (Gunstad et al., 2008; Lommatzsch et al.,

2005; Ziegenhorn et al., 2007). Ziegenhorn et al. (2007) found serum BDNF levels were negatively correlated with age in healthy old adults ranging in age from 70 to 103 years (i.e. levels decreased with increasing age). Bringing all of this evidence together, Erickson et al., (2010) also found increasing age was associated with reduced levels of BDNF, and further that reduced levels of BDNF were related to both a decline in HC volume and elevated memory deficits. xxx

In a rodent model of successful aging (i.e. longer lifespans and preserved memory capacities), BDNF levels were higher than compared to those experiencing normal age-related patterns of decline (Silhol et al., 2008). This is evidence that natural genetic variation can contribute to successful aging. But is there anything that can be done for those with naturally lower levels of BDNF? Yes, induction of BDNF production and secretion in the HC has been shown to rescue LTP and relieve spatial memory deficits in aged mice (Rex et al., 2006, 2007; Simmons et al., 2009). So is there an easy solution? Again, the answer is yes. Exercise has been shown to increase BDNF levels in the rat hippocampus, cerebellum and frontal cortex, most likely through increased gene expression (Ding et al., 2006; Neeper et al., 1995). In fact, Vaynman, Ying, & Gomez-Pinilla (2004) found that inhibiting BDNF action blocked the beneficial effects of exercise on performance of the Morris water maze in rats, indicating that BDNF may be one of the mediators of exercise-induced cognitive enhancement (Ding et al., 2006; Vaynman et al., 2004). Further, regular physical exercise in rats, specifically wheel-running, stimulates brain vascularization (Kleim, Cooper, & VandenBerg, 2002), increases levels in brain catecholamines, particularly dopamine and noradrenaline (Sutoo & Akiyama, 2003), and BDNF, which in turn increases neuronal survival (Berchtold, Castello, & Cotman, 2010; Barde, 1994) and neurogenesis (Creer et al., 2010; van Praag, Christie, et al., 1999; van Praag, Kempermann, et al., 1999).

In summary, animal studies suggest BDNF protects neurons from effects of damage, plays a role in modifying synaptic connections, modulates hippocampal LTP, and can influence behavior – specifically memory. BDNF levels are influenced by age, exercise, and under some level of genetic control. Studies in human subjects demonstrate that the Met allele of the BDNF Val66Met SNP is associated with lower hippocampal volume, functional activity, and with weaker performance on tests of memory and executive function.

BDNF Val66Met (rs6265) Predictions

The extant literature suggests that Met Carriers have diminished activity-dependent secretion of BDNF and reduced performance on behavioral tests. Therefore, I expect that in my sample, the Met Carriers will also show reduced cognitive performance compared to non-Met Carriers. In terms of the interaction between BDNF Val66Met and cardiorespiratory fitness level, I predict a stronger positive relationship between Fitness Level and behavioral performance in Met Carriers compared to non-Met Carriers. This prediction is based on the assumption that Met Carriers will benefit cognitively from a fitness-induced increase of BDNF.

BDNF Val66Met (rs6265) Results

There were no significant differences between the groups on age, education, mMMSE, BMI or VO_2 (Table 4.6). Chi-square analysis showed the distribution of males and females was not significantly difference between the groups (Table 4.6). Task means and standard deviations are reported in Table 4.7.

BDNF: Verbal Crystallized Intelligence

Verbal crystallized intelligence has been shown to remain relatively stable in older adult populations (Hedden & Gabrieli, 2004; Park & Gutchess, 2002). Due to the relative stability of the verbal crystallized intelligence, I do not expect to find an effect of the BDNF Val66Met SNP or its interaction with cardiorespiratory fitness level.

KBIT test. The sample size for the KBIT test was $N = 194$, with group sizes as follows: Val/Val = 177, Met Carriers = 17. Analysis of covariance (ANCOVA) showed non-significant differences between the BDNF Val66Met allelic groups, $F(1, 189) < 1$.

When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 159$, with allelic group sizes as follows: Val/Val = 144, Met Carriers = 15. The interaction between BDNF Val66Met X Fitness Level explained 2.8% of the variance in age-scaled scores, $\Delta R^2 = .028$, $\Delta F(1, 152) = 5.56$, $p = .020$ (Figure 4.10). Simple slopes analysis revealed the age-scaled scores increased with increasing VO_2 for the Met Carriers, $B = 2.22$, $\beta = 1.31$, $t(152) = 2.64$, $p = .002$, 95% CI [-0.10, 3.31], but not for Val/Val

Homozygotes, $B = 0.20$, $\beta = 0.15$, $t(152) = 0.94$, $p = .372$, 95% CI [-0.23, 0.64]. These results suggest verbal crystallized intelligence can be differentially influenced by varying cardiorespiratory fitness level and BDNF secretion.

BDNF: Memory Tests

Performance on tests of working and spatial memory are seen to decrease in older adult populations, in contrast to more stable performance on tests of short-term memory (Hedden & Gabrieli, 2004; Park & Gutchess, 2002). In terms of the BDNF Val66Met SNP, Met Carriers are biologically associated with less BDNF secretion and have been shown to have a reduced performance compared to Val/Val homozygotes on tests of working and spatial memory, but not short-term memory (Egan et al., 2003; Hariri et al., 2003). In terms of the BDNF Val66Met allele, Met Carriers have been shown to have a reduced performance compared to Val/Val homozygotes on tests of working and spatial memory, but not short-term memory (Egan et al., 2003; Hariri et al., 2003). The extant literature shows that memory performance increases with increasing fitness levels. Taking into consideration these previous findings, I expect that Met Carriers will show a greater behavioral benefit from increasing cardiorespiratory fitness levels compared to Val/Val homozygotes.

Digit span test. The sample size was $N = 196$ with allelic group sizes as follows: Val/Val = 179, Met Carriers = 17. There were no significant group differences on the number of digits repeated for either version of the digit span test, forward and backward both $F(1, 191) < 1$. When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 161$ with allelic group sizes as follows: Val/Val = 146, Met Carriers = 15. The interaction did not significantly explain variance in either version: forward, $\Delta R^2 = .000$, $\Delta F(1, 154) < 1$; backward, $\Delta R^2 = .001$, $\Delta F(1, 154) < 1$.

Spatial memory test. The sample size was $N = 176$ with allelic group sizes as follows: Val/Val = 160, Met Carriers = 16. There were no significant group differences on response time (RT) or error rate (ER) for any memory load (Table B.3). When cardiorespiratory fitness level was added into the model, the sample size was reduced by $N = 144$ with allelic group sizes as follows: Val/Val = 130, Met Carriers = 14. The interaction

between BDNF Val66Met X Fitness Level did not significantly explain variance in RT or ER for any memory load (Table 4.8).

BDNF: Executive Control Function Tests

Increasing age most negatively affects performances on tests requiring some form of executive control (Hedden & Gabrieli, 2004; Park & Gutchess, 2002). I expect Val/Val homozygotes to perform better than Met allele carriers. Increasing age most negatively affects performances on tests requiring some form of executive control (Hedden & Gabrieli, 2004; Park & Gutchess, 2002), but higher levels of fitness tend to be associated with better executive control (Colcombe & Kramer, 2003). The Met allele is biologically associated with less BDNF secretion and behaviorally with worse performance on tests of executive control (Egan et al., 2003). Therefore, I expect Val/Val homozygotes to outperform carriers of the Met allele, and an interaction effect such that Met Carriers will show a positive fitness effect, but the Val/Val homozygotes will show less of an effect or no effect at all.

Verbal fluency test (FAS). The sample size for the word fluency test was $N = 196$, with group sizes as follows: Met Carriers = 17, Val/Val = 179. ANCOVA showed no differences between the allelic groups on words named, $F(1, 191) < 1$. When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 161$, with group sizes as follows: Met Carriers = 15, Val/Val = 146. Hierarchical regression showed the interaction between BDNF Val66Met x Fitness Level marginally explained 1.7% of the variance in the number of words named, $\Delta R^2 = .017$, $\Delta F(1, 154) = 2.98$, $p = .086$ (Figure 4.8). Simple slopes analysis revealed the number of words named marginally increased with increasing VO_2 for the Met Carriers, $B = 1.70$, $\beta = 0.98$, $t(154) = 1.84$, $p = .074$, 95% CI [-0.70, 3.67], but not for the Val/Val Homozygotes, $B = 0.08$, $\beta = 0.06$, $t(154) = 0.34$, $p = .706$, 95% CI [-0.32, 0.53].

Wisconsin card sorting test (WCST). The sample size for the WCST was $N = 182$, with group sizes as follows: Met Carriers = 14, Val/Val = 168. ANCOVA did not show significant difference between the groups on performance measured by total errors, $F(1, 177) = 1.68$, $p = .196$, or percent perseverative errors, $F(1, 177) < 1$. When cardiorespiratory fitness level was added into the model, the sample size was reduced to N

= 150, with group sizes as follows: Met Carriers = 13, Val/Val = 137. Hierarchical regression showed the interaction between BDNF Val66Met x Fitness Level did not significantly explain any variance in total errors, $\Delta R^2 = .007$, $\Delta F(1, 143) = 1.09$, $p = .298$, but it did marginally explain 2.2% of the variance in percent perseverative errors, $\Delta R^2 = .022$, $\Delta F(1, 143) = 3.44$, $p = .066$ (Figure 4.12). Simple slopes analysis revealed percent perseverative errors increased with increasing VO_2 for the Met Carriers, $B = 1.26$, $\beta = 0.87$, $t(143) = 1.52$, $p = .029$, 95% CI [0.20, 3.08], but not for the Val/Val Homozygotes, $B = -0.30$, $\beta = -0.26$, $t(143) = -1.55$, $p = .134$, 95% CI [-0.71, 0.07]. This was unexpected since higher levels of cardiorespiratory fitness are typically associated with increased cognitive flexibility. These results suggest a more complex relationship between fitness and behavior overall, and further when other factors, such as the BDNF Val66Met SNP, are taken into account.

Dual task test. The sample size for the dual task test $N = 158$, with group sizes as follows: Met Carriers = 15, Val/Val = 143. ANCOVA showed no differences between the groups on response time (RT) for either single, $F(1, 153) < 1$, or dual trials, $F(1, 153) = 1.59$, $p = .209$ (Figure B.5a). However, the groups did significantly differ on an RT cost score (i.e. dual RT – single RT), $F(1, 153) = 4.34$, $p = .039$, with the Met Carriers showing a lower RT cost compared to the Val/Val Homozygotes, mean difference = -69.12, $p = .011$, 95% CI [-120.74, -13.84] (Figure B.5b). Further, there were no differences between the groups on error rate (ER) for either trial type, both $F(1, 153) < 1$. When cardiorespiratory fitness level was added into the model, the sample size was reduced to $N = 151$, with group sizes as follows: Met Carriers = 14, Val/Val = 137. Hierarchical regression showed that the interaction between BDNF Val66Met x Fitness Level did not significantly explain any variance in RT or ER for either trial type (Table 4.9).

Growth Factors Combined

To further study the effects of the growth factor genes on cognition in healthy older adults, I conducted an analysis using a combination of the two SNPs. I created a high growth factor (high GF; i.e. high IGF1 and BDNF) group which included those that were Val/Val homozygotes on the BDNF Val66Met SNP and G/G homozygotes on the IGF1 G/A SNP. I compared this group to a low growth factor (low GF; i.e. low IGF1 and BDNF) group

which included those that were Met allele carriers on the BDNF Val66Met SNP and A/A homozygotes on the IGF1 G/A SNP.

There were no significant differences between the Low GF and High GF groups on education, mMMSE, BMI or VO_2, but there was a marginal age difference (Table 4.10). Chi-square analysis showed the low GF group had less males and more females than expected and vice versa for the high GF group. Means and standard deviations for the primary measures on the behavioral tests are reported in in Table 4.11.

Verbal Crystallized Intelligence

KBIT test. The sample size for the KBIT test was $N = 30$, with genotypic group sizes as follows: low GF = 10, high GF = 20. As expected, there was no significant difference between the groups on the age-scaled scores $F(1, 25) < 1$, suggesting the growth factor genes does not influence verbal crystallized intelligence.

When cardiorespiratory fitness level was added to the model, the sample size for the KBIT test was reduced to $N = 27$, with genotypic group sizes as follows: low GF = 8, high GF = 19. Hierarchical regression showed the interaction between GF Genes x Fitness Level did not explain a significant proportion of variance in the age-scaled scores, $\Delta R^2 = .024$, $\Delta F(1, 20) = 1.01$, $p = .328$. This followed predictions that the growth factor genes nor cardiorespiratory fitness level would affect verbal crystallized intelligence.

Memory Tests

Digit span test. The sample size for the digit span test was $N = 30$, with genotypic group sizes as follows: low GF = 10, high GF = 20. No significant differences were found between the groups for forward span, $F(1, 25) < 1$, but the low GF group performed significantly better on the backward span, $F(1, 25) = 4.60$, $p = .042$, mean difference = 1.09, $p = .042$, 95% CI [-0.04, 1.99] (Figure B.6). These results were expected for the forward span, a test of short-term memory, but for the backward span, a test of working memory, I did not expect the low GF group to perform better than the high GF group.

When cardiorespiratory fitness level was added into the model the sample size for the digit span tests was reduced to $N = 27$, with genotypic group sizes as follows: low GF = 8, high GF = 19. Hierarchical regression showed no interaction effect of GF Genes x Fitness

Level on either forward span, $\Delta R^2 = .068$, $\Delta F(1, 20) = 1.71$, $p = .205$, or backward span, $\Delta R^2 = .014$, $\Delta F(1, 20) < 1$. Based on previous studies showing increasing fitness levels can increase GF levels, and that the GF genotypes influences GF levels, I expected a potential interaction between the two variables.

Spatial memory test. The sample size for the spatial memory test was $N = 23$ with genotypic group sizes as follows: low GF = 9, high GF = 14. Against prediction, there were no significant differences between the groups on either response time (RT) or error rate (ER) for any memory load (Table B.3).

Addition of cardiorespiratory fitness level reduced the sample size to $N = 20$, with genotypic group sizes as follows: low GF = 7, high GF = 13. The interaction between GF Genes x Fitness Level did not significantly explain variance in RT or ER for any memory load (Table 4.12).

Executive Control Function Tests

Verbal fluency test (FAS). The sample size was $N = 30$, with genotypic group sizes as follows: low GF = 10, high GF = 20. There were no significant group differences on the number of words named, $F(1, 25) < 1$. When cardiorespiratory fitness level was added to the model, the sample size was reduced to $N = 27$, with genotypic group sizes as follows: low GF = 8, high GF = 19. The interaction between the GF Genes and Fitness significantly explained 11.3% of the variance in performance of the verbal fluency test, $\Delta R^2 = .113$, $\Delta F(1, 20) = 5.74$, $p = .026$ (Figure 4.10). Simple slopes analysis showed the number of words named significantly increased with VO_2 only for the low GF group, $B = 2.92$, $\beta = 2.65$, $t(20) = 3.09$, $p = .014$, 95% CI [-0.80, 5.86], but not the high GF group, $B = 0.55$, $\beta = 0.50$, $t(20) = 1.04$, $p = .408$, 95% CI [-0.48, 1.78].

Growth Factor Tables

Table 4.1 Demographics and physical assessment descriptive statistics for IGF1 G/A (rs6220)

	A/A		A/G		G/G			Total	
	n = 103 (35 m)		*n* = 72 (30 m)		*n* = 20 (9 m)			*N* = 195 (74 m)	
Demographics	**Mean**	***SD***	**Mean**	***SD***	**Mean**	***SD***	***F*(2, 192)**	**Mean**	***SD***
Age	65.92	(5.05)	66.72	(6.54)	66.95	(5.30)	0.56	66.32	(5.65)
Educuation (years)	15.67	(3.28)	16.22	(2.94)	15.75	(2.71)	0.69	15.88	(3.10)
mMMSE	54.53	(2.41)	54.86	(2.37)	54.10	(2.83)	0.87	54.61	(2.44)

mMMSE = modified mini mental status exam; m = males χ^2 (2, N =195) = 1.53, p = .464

	A/A		A/G		G/G			Total	
	n = 28 (11 m)		*n* = 87 (28 m)		*n* = 46 (19 m)			*N* = 161 (58 m)	
Physical Assessment	**Mean**	***SD***	**Mean**	***SD***	**Mean**	***SD***	***F*(2, 158)**	**Mean**	***SD***
Body mass index (kg · m	28.99	(4.54)	28.64	(4.32)	29.58	(3.15)	0.22	28.93	(4.30)
VO2 (mL · kg $^{-1}$ · min $^{-1}$)	21.33	(4.83)	21.38	(4.38)	19.96	(5.40)	1.21	21.19	(4.73)

m = males χ^2 (2, N =161) = 1.52 p = .468

Table 4.2 IGF1 G/A (rs6220) means and standard deviations

	A/A		A/G		G/G		Total	
	Mean	(*SD*)	Mean	(*SD*)	Mean	(*SD*)	Mean	(*SD*)
Verbal Crystallized Intelligence								
KBIT	112.43	(9.46)	112.24	(10.61)	111.95	(13.84)	112.31	(10.36)
Memory Tests								
Digit Span								
Forward	6.38	(1.14)	6.69	(1.22)	6.40	(1.05)	6.50	(1.17)
Backward	4.95	(1.47)	4.99	(1.32)	4.60	(1.19)	4.93	(1.39)
Spatial Memory								
Load 1 RT (ms)	816.91	(176.70)	754.97	(182.65)	835.54	(186.43)	794.69	(181.51)
Load 2 RT (ms)	910.90	(167.39)	871.78	(189.62)	916.04	(185.63)	896.33	(177.67)
Load 3 RT (ms)	1011.07	(179.09)	935.49	(203.48)	1034.86	(193.77)	984.04	(198.78)
Load 1 Error Rate	0.10	(0.08)	0.11	(0.10)	0.11	(0.06)	0.10	(0.08)
Load 2 Error Rate	0.15	(0.09)	0.16	(0.11)	0.18	(0.10)	0.16	(0.10)
Load 3 Error Rate	0.20	(0.10)	0.20	(0.12)	0.18	(0.11)	0.20	(0.11)
Executive Control Function Tests								
Stroop								
Neutral RT (ms)	809.88	(89.59)	794.24	(100.50)	769.23	(68.73)	799.34	(92.23)
Congruent RT (ms)	800.02	(100.79)	781.30	(90.47)	779.92	(65.51)	790.68	(93.52)
Ineligible Incongruent RT (ms)	866.70	(104.37)	836.81	(106.42)	816.19	(80.17)	849.68	(103.77)
Eligible Incongruent RT (ms)	894.55	(114.93)	871.40	(116.83)	853.19	(100.56)	881.11	(114.39)
Neutral Error Rate	0.04	(0.05)	0.04	(0.06)	0.03	(0.05)	0.04	(0.06)
Congruent Error Rate	0.04	(0.05)	0.04	(0.08)	0.02	(0.04)	0.04	(0.06)
Ineligible Incongruent Error Rate	0.06	(0.07)	0.06	(0.09)	0.03	(0.03)	0.06	(0.08)
Eligible Incongruent Error Rate	0.07	(0.07)	0.08	(0.11)	0.05	(0.06)	0.07	(0.09)
Task Switch								
Single Condition RT (ms)	777.21	(114.59)	752.86	(104.45)	789.33	(142.41)	770.07	(114.66)
Mix Condition RT (ms)	1152.61	(156.57)	1135.48	(159.55)	1196.28	(125.92)	1151.61	(154.61)
Global RT Cost (ms)	375.40	(128.15)	382.61	(134.22)	406.94	(126.99)	381.54	(129.71)
Repeat Trial RT (ms)	987.23	(143.30)	950.96	(156.46)	990.19	(132.00)	974.87	(147.00)
Switch Trial RT (ms)	1317.99	(199.42)	1319.99	(209.79)	1402.36	(160.87)	1328.36	(199.82)
Local Cost (ms)	330.76	(150.15)	369.03	(187.50)	412.17	(152.27)	353.50	(165.69)
Single Condition Error Rate	0.05	(0.07)	0.06	(0.08)	0.10	(0.14)	0.06	(0.08)
Mix Condition Error Rate	0.12	(0.11)	0.10	(0.11)	0.17	(0.10)	0.12	(0.11)
Global Error Rate Cost	0.06	(0.10)	0.04	(0.10)	0.06	(0.15)	0.05	(0.11)
Repeat Error Rate	0.10	(0.10)	0.08	(0.01)	0.13	(0.09)	0.10	(0.10)
Switch Error Rate	0.13	(0.12)	0.12	(0.12)	0.20	(0.12)	0.14	(0.12)
Local Error Rate Cost	0.03	(0.06)	0.04	(0.05)	0.07	(0.07)	0.04	(0.06)
Dual Task								
Single RT (ms)	1222.75	(166.68)	1231.09	(167.73)	1231.22	(147.97)	1226.88	(164.10)
Dual RT (ms)	2035.30	(208.61)	2005.18	(189.58)	1961.33	(188.56)	2015.38	(199.57)
RT Cost (ms)	812.55	(131.77)	774.10	(113.34)	730.12	(135.54)	788.50	(127.78)
Single Error Rate	0.05	(0.06)	0.08	(0.09)	0.04	(0.07)	0.06	(0.07)
Dual Error Rate	0.14	(0.13)	0.16	(0.16)	0.09	(0.07)	0.14	(0.14)
Error Rate Cost (ms)	0.09	(0.11)	0.08	(0.10)	0.05	(0.06)	0.08	(0.10)

Table 4.3 Summary of hierarchical regression analysis of the effect of the interaction between IGF1 G/A x Fitness Level on performance of the spatial memory test

	ΔR^2	$\Delta F(2, 135)$	p
Memory Load 1			
Response Time	0.009	0.715	.491
Error Rate	0.010	0.72	.488
Memory Load 2			
Response Time	0.019	1.475	.232
Error Rate	0.047*	3.555	.031
Memory Load 3			
Response Time	0.026	2.056	.132
Error Rate	0.001	0.103	.902

* $p \leq .05$

Table 4.4 Summary of hierarchical regression analysis of the effect of the interaction between IGF1 G/A x Fitness Level on performance of the Stroop test

	ΔR^2	$\Delta F(2, 142)$	p
Neutral Trial			
Response Time	0.066*	5.500	.005
Error Rate	0.013	1.060	.349
Congruent Trial			
Response Time	0.041*	3.290	.040
Error Rate	0.012	0.955	.387
Incongruent Ineligible Trial			
Response Time	0.040*	3.270	.041
Error Rate	0.020	1.620	.201
Incongruent Eligible Trial			
Response Time	0.038*	3.080	.049
Error Rate	0.033 +	2.830	.062

Table 4.5 Summary of simple slopes analysis for the effect of the interaction between IGF1 G/A x Fitness Level on performance of the Stroop test

					95% Confidence Interval	
	B	β	*t* (142)	*p*	Lower	Upper
Response Time						
Neutral Trial						
A/A	0.087	0.011	0.038	.968	-4.450	4.628
A/G	-9.149*	-1.144	-3.161	.002	-15.313	-3.576
G/G	0.388	0.030	0.088	.921	-9.354	7.085
Congruent Trial						
A/A	0.315	0.042	0.141	.888	-4.391	4.799
A/G	-7.014*	-0.911	-2.475	.028	-13.832	-1.322
G/G	-3.877	-3.150	-0.902	.210	-11.480	2.559
Incongruent Ineligible Trial						
A/A	-0.815	-0.089	-0.300	.784	-6.724	4.727
A/G	-8.312*	-0.881	-2.423	.012	-14.847	-1.422
G/G	2.167	0.144	0.417	.582	-6.523	10.448
Incongruent Eligible Trial						
A/A	-0.280	-0.029	-0.095	.929	-6.613	5.329
A/G	-8.976*	-0.881	-2.401	.016	-16.894	-2.010
G/G	2.146	0.132	0.379	.662	-10.653	10.699

* $p \leq .05$

Table 4.6 Demographics and physical assessment descriptive statistics for BDNF Val66Met (rs6265)

	Val/Val (G/G)		**Met (A) Allele**			**Total**	
	***n* = 180 (70 m)**		***n* = 17 (5 m)**			***N* = 197 (75 m)**	
Demographics	**Mean**	***SD***	**Mean**	***SD***	***F*(1, 195)**	**Mean**	***SD***
Age	66.58	(5.78)	64.88	(5.07)	1.36	66.43	(5.73)
Educuation (years)	15.98	(3.06)	14.88	(3.26)	1.97	15.88	(3.08)
mMMSE	54.59	(2.46)	54.71	(2.17)	0.04	54.60	(2.43)

MMSE = modified mini mental status exam; m = males χ^2 (1, *N* =197) = .592, *p* = .442

	Val/Val (G/G)		**Met (A) Allele**			**Total**	
	***n* = 147 (54 m)**		***n* = 15 (4 m)**			***N* = 162 (58 m)**	
Physical Assessment	**Mean**	***SD***	**Mean**	***SD***	***F*(1, 160)**	**Mean**	***SD***
Body mass index ($kg \cdot m^{-2}$)	29.05	(4.40)	27.33	(3.13)	2.25	28.89	(4.32)
VO2 ($mL \cdot kg^{-1} \cdot min^{-1}$)	21.23	(4.85)	20.85	(3.08)	0.01	21.19	(4.71)

m = males χ^2 (1, *N* =162) = .600 *p* = .438

Table 4.7 BDNF Val66Met (rs6265) means and stanadard deviations

	Val/Val (G/G)		Met (A) Allele		Total	
	Mean	(*SD*)	Mean	(*SD*)	Mean	(*SD*)
Verbal Crystallized Intelligence						
KBIT	112.49	(10.50)	111.71	(9.00)	112.42	(10.36)
Memory Tests						
Digit Span						
Forward	6.53	(0.18)	6.24	(0.97)	6.50	(1.16)
Backward	4.90	(1.39)	5.12	(1.36)	4.92	(1.39)
Spatial Memory						
Load 1 RT (ms)	797.46	(182.87)	769.56	(164.52)	794.92	(181.02)
Load 2 RT (ms)	801.28	(177.39)	855.72	(178.61)	897.13	(177.48)
Load 3 RT (ms)	988.76	(192.88)	942.19	(187.37)	984.52	(192.33)
Load 1 Error Rate	0.11	(0.09)	0.08	(0.06)	0.10	(0.08)
Load 2 Error Rate	0.16	(0.10)	0.12	(0.07)	0.16	(0.10)
Load 3 Error Rate	0.20	(0.11)	0.19	(0.09)	0.20	(0.11)
Executive Control Function Tests						
FAS	37.27	(10.88)	38.71	(10.22)	37.39	(10.81)
WCST						
Total Errors	42.90	(23.91)	95.07	(17.74)	42.30	(23.54)
Percent Perseverative Errors	17.64	(9.52)	14.64	(6.33)	17.41	(9.34)
Dual Task						
Single RT (ms)	1225.91	(166.06)	1223.37	(149.34)	1225.67	(164.10)
Dual RT (ms)	2021.85	(200.34)	1952.34	(176.11)	2015.25	(198.70)
RT Cost (ms)	795.94	(127.51)	728.97	(112.35)	789.58	(127.36)
Single Error Rate	0.06	(0.08)	0.07	(0.08)	0.06	(0.08)
Dual Error Rate	0.14	(0.14)	0.13	(0.11)	0.14	(0.14)
Error Rate Cost (ms)	0.08	(0.10)	0.06	(0.06)	0.08	(0.10)

Table 4.8 Summary of hierarchical regression analysis of the effect of the interaction between BDNF Val66Met x Fitness Level on performance of the spatial memory test

	ΔR^2	$\Delta F(1, 137)$	p
Memory Load 1			
Response Time	0.001	0.077	.781
Error Rate	0.003	0.500	.481
Memory Load 2			
Response Time	0.000	0.000	.990
Error Rate	0.014	2.126	.147
Memory Load 3			
Response Time	0.001	0.145	.704
Error Rate	0.011	1.859	.175

Table 4.9 Summary of hierarchical regression analysis of the effect of the interaction between BDNF Val66Met x Fitness Level on performance of the dual task test

	ΔR^2	$\Delta F(1, 137)$	p
Single Trial Type			
Response Time	0.006	0.926	.338
Error Rate	0.000	0.002	.960
Dual Trial Type			
Response Time	0.000	0.057	.812
Error Rate	0.001	0.176	.676

Table 4.10 Demographics and physical assessment descriptive statistics for growth factors (GF) combined

	Low GF		High GF			Total	
	n = 10 (1 m)		*n* = 20 (9 m)			*N* = 30 (10 m)	
Demographics	**Mean**	***SD***	**Mean**	***SD***	***F* (1, 28)**	**Mean**	***SD***
Age	63.60	(4.35)	66.95	(5.30)	2.98 +	65.83	(5.18)
Educuation (years)	14.90	(4.12)	15.75	(2.71)	0.46	15.47	(3.20)
mMMSE	54.40	(2.37)	54.10	(2.83)	0.08	54.20	(2.64)

MMSE = modified mini mental status exam; m = males; + χ^2 (1, *N* =30) = 3.68, *p* = .055

	Low GF		High GF			Total	
	n = 8 (0 m)		*n* = 19 (8 m)			*N* = 27 (8 m)	
Physical Assessment	**Mean**	***SD***	**Mean**	***SD***	***F* (1, 25)**	**Mean**	***SD***
Body mass index (kg · m	28.13	(3.48)	29.58	(3.15)	1.13	29.15	(3.25)
VO2 (mL · kg $^{-1}$ · min $^{-}$	20.85	(3.46)	19.96	(5.40)	0.18	20.23	(4.86)

*m = males; * p ≤ .05* * χ^2 (1, *N* =27) = 4.79 *p* = .029

Table 4.11 Growth factor genes combined means and stanadard deviations

	Low GF (Met Allele + A/A)		High GF (Val/Val + G/G)		Total	
	Mean	**(*SD*)**	**Mean**	**(*SD*)**	**Mean**	**(*SD*)**
Verbal Crystallized Intelligence						
KBIT	109.80	(9.21)	111.95	(13.84)	111.23	(12.36)
Memory Tests						
Digit Span						
Forward	6.30	(1.16)	6.40	(1.05)	6.37	(1.07)
Backward	5.50	(1.51)	4.60	(1.19)	4.90	(1.35)
Spatial Memory						
Load 1 RT (ms)	804.33	(115.02)	835.54	(186.43)	823.33	######
Load 2 RT (ms)	878.17	(143.47)	916.04	(185.63)	901.22	######
Load 3 RT (ms)	964.56	(146.36)	1034.86	(193.77)	######	######
Load 1 Error Rate	0.07	(0.03)	0.11	(0.06)	0.10	(0.05)
Load 2 Error Rate	0.11	(0.07)	0.18	(0.10)	0.15	(0.09)
Load 3 Error Rate	0.17	(0.05)	0.18	(0.11)	0.18	(0.09)
Executive Control Function Tests						
FAS	38.20	(12.87)	34.80	(11.25)	35.93	(11.70)

Table 4.12 Summary of hierarchical regression analysis of the effect of the interaction between Growth Factor Genes x Fitness Level on performance of the spatial memory test

	ΔR^2	$\Delta F(1, 13)$	p
Memory Load 1			
Response Time	0.000	0.000	0.991
Error Rate	0.002	0.038	0.848
Memory Load 2			
Response Time	0.008	0.121	0.734
Error Rate	0.012	0.218	0.648
Memory Load 3			
Response Time	0.009	0.137	0.717
Error Rate	0.103	1.892	0.192

Growth Factor Figures

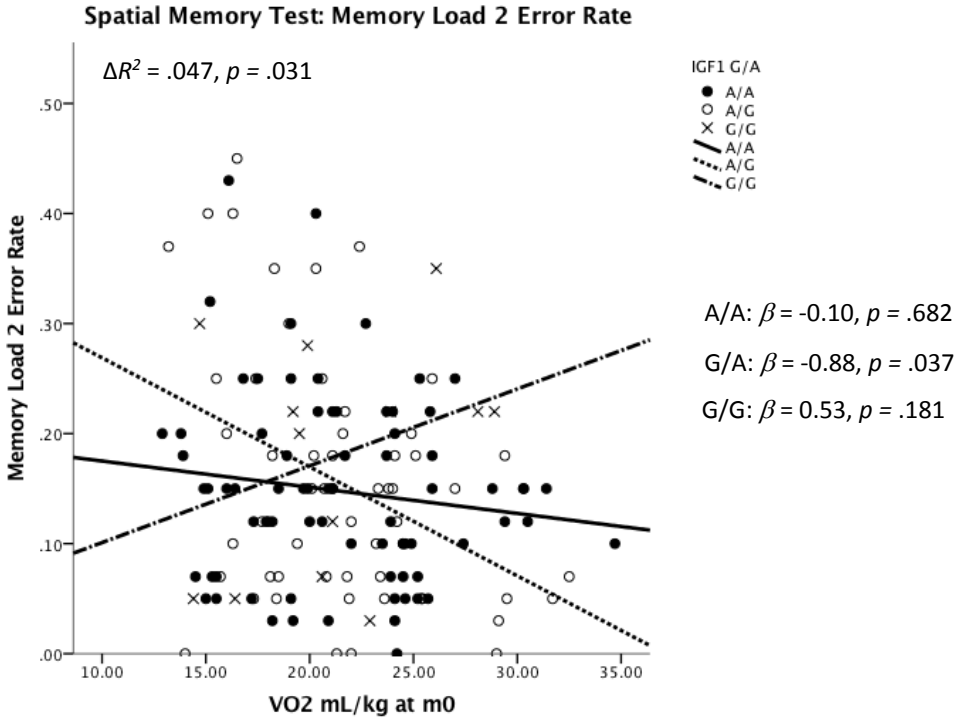


Figure 4.1 Interaction effect between the IGF1 G/A SNP and cardiorespiratory fitness level (i.e. VO_2) on error rate for memory load 2 of the spatial memory test. The interaction significantly explained 4.7% of the variation in error rate for memory load 2, $\Delta R^2 = .047$, $\Delta F(2, 135) = 3.56$, $p = .031$. Simple slopes A/A, $B = -.001$, $\beta = -0.10$, $t(135) = -0.33$, $p = .682$, 95% CI [-.005, .003]; G/A group, $B = -.008$, $\beta = -0.88$, $t(135) = -2.40$, $p = .037$, 95% CI [-.015, .000]; G/G, $B = .008$, $\beta = 0.53$, $t(135) = 1.37$, $p = .181$, 95% CI [-.007, .022].

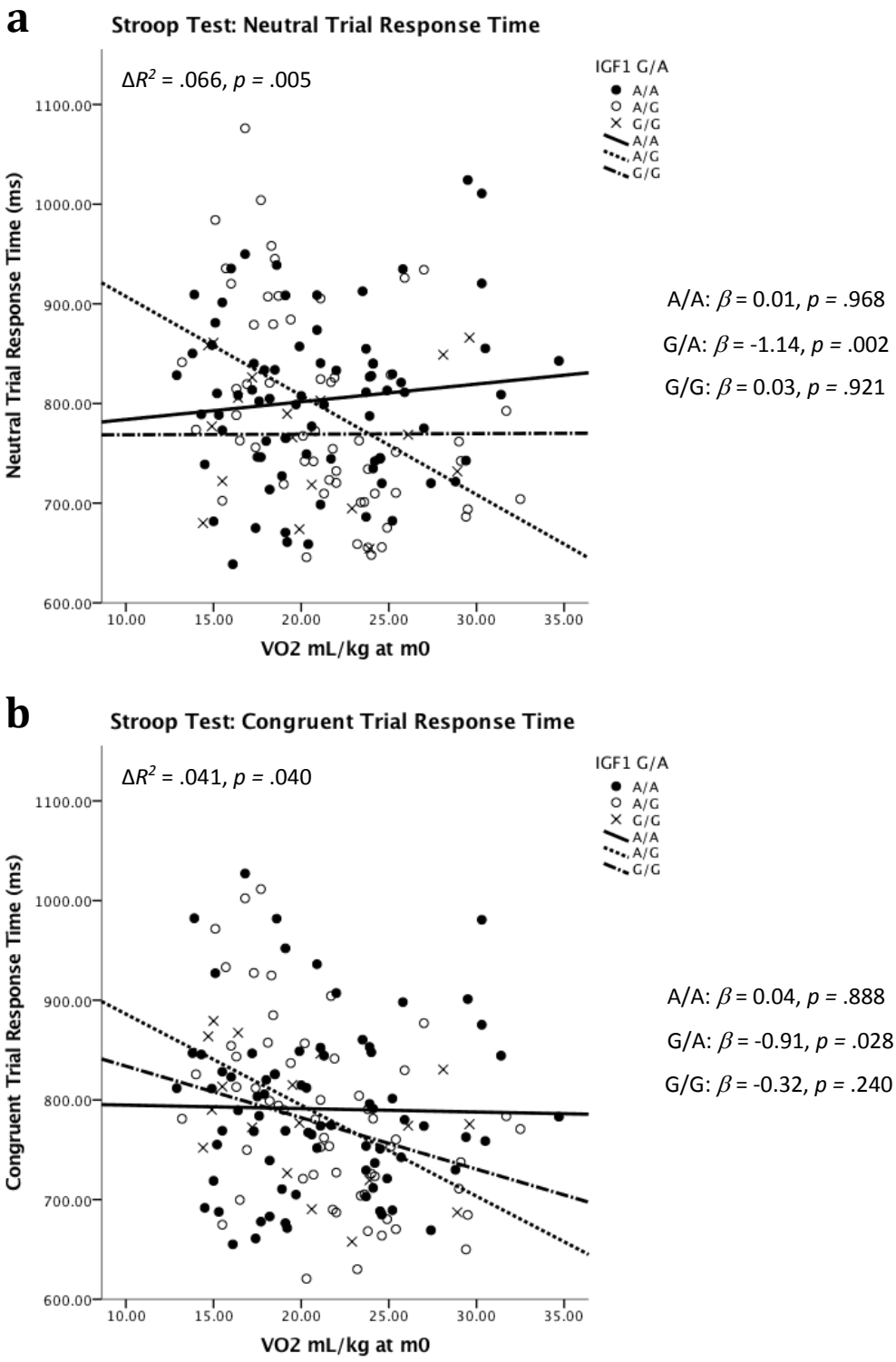
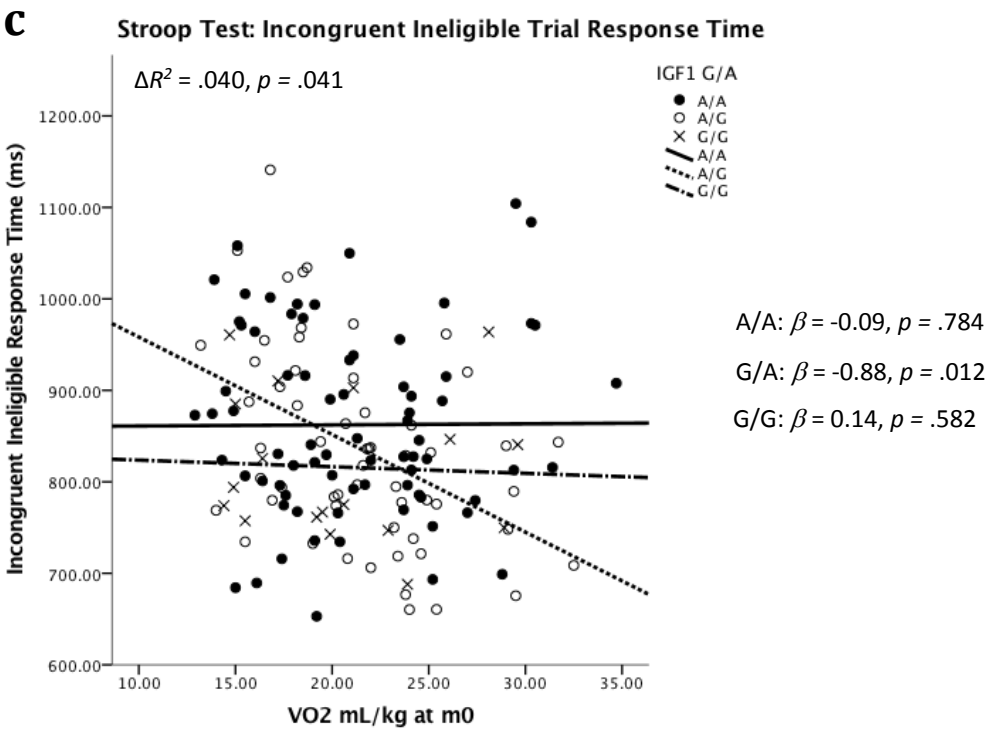


Figure 4.2 Interaction effect between the IGF1 G/A SNP and cardiorespiratory fitness level (i.e. VO_2) on response time for all trial types of the Stroop test. The interaction between IGF1 G/A x Fitness Level significantly explained variance in response time for each trial type (simple slopes reported in Table 4.5) **a)** neutral RT, $\Delta R^2 = .066$, $\Delta F(2, 142) = 5.50$, $p = .005$ **b)** congruent RT, $\Delta R^2 = .041$, $\Delta F(2, 142) = 3.29$, $p = .040$



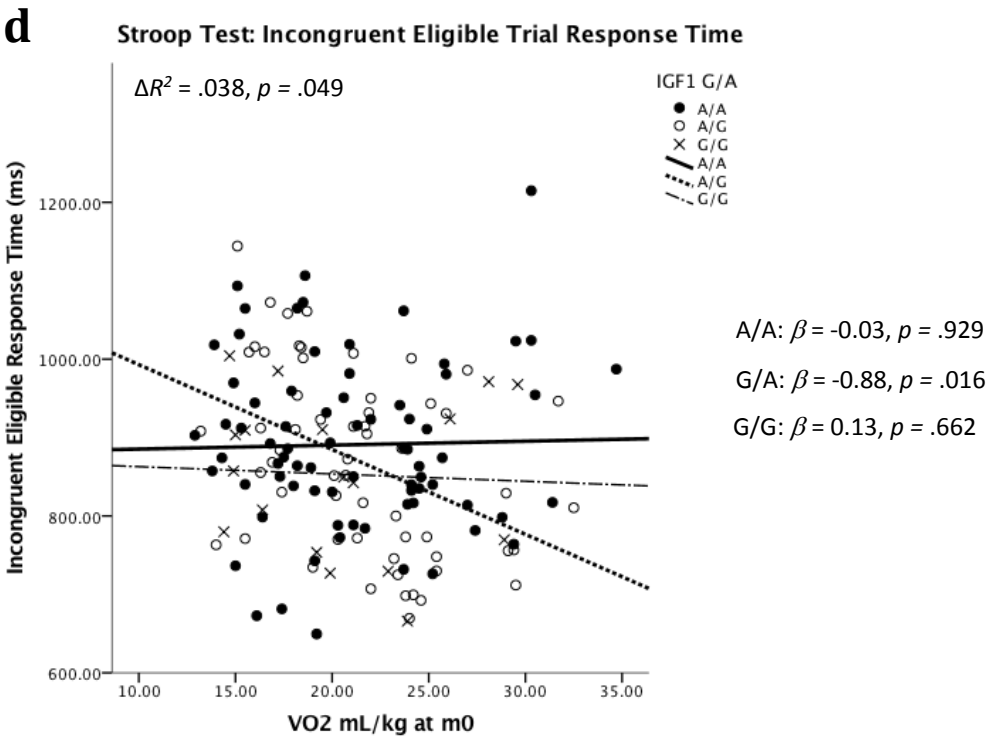


Figure 4.2 cont. Interaction effect between the IGF1 G/A SNP and cardiorespiratory fitness level (i.e. VO_2) on response time for all trial types of the Stroop test. The interaction between IGF1 G/A x Fitness Level significantly explained variance in response time for each trial type (simple slopes reported in Table 4.5) **c)** incongruent-ineligible RT, $\Delta R^2 = .040$, $\Delta F(2, 142) = 3.28$, $p = .041$ **d)** incongruent eligible RT, $\Delta R^2 = .038$, $\Delta F(2, 142) = 3.08$, $p = .049$.

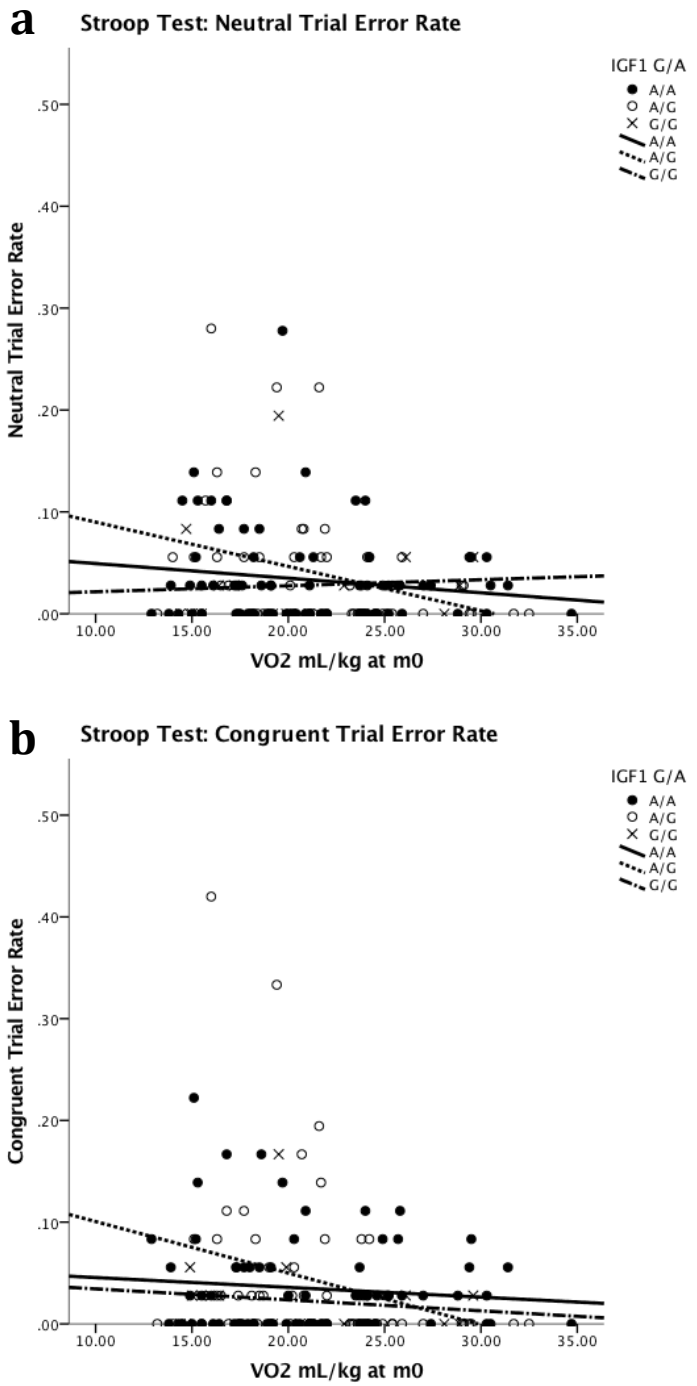


Figure 4.3 Interaction effect between the IGF1 G/A SNP and cardiorespiratory fitness level (i.e. VO_2) on error rate for all trial types of the Stroop test. The interaction between IGF1 G/A x Fitness Level marginally explained variance in error rate only for the incongruent eligible trials but the trend was similarly for other trial types **a)** neutral RT, $\Delta R^2 = 0.01$, $\Delta F(2, 142) = 1.06$, $p = .349$ **b)** congruent RT, $\Delta R^2 = 0.01$, $\Delta F(2, 142) = 0.96$, $p = .387$

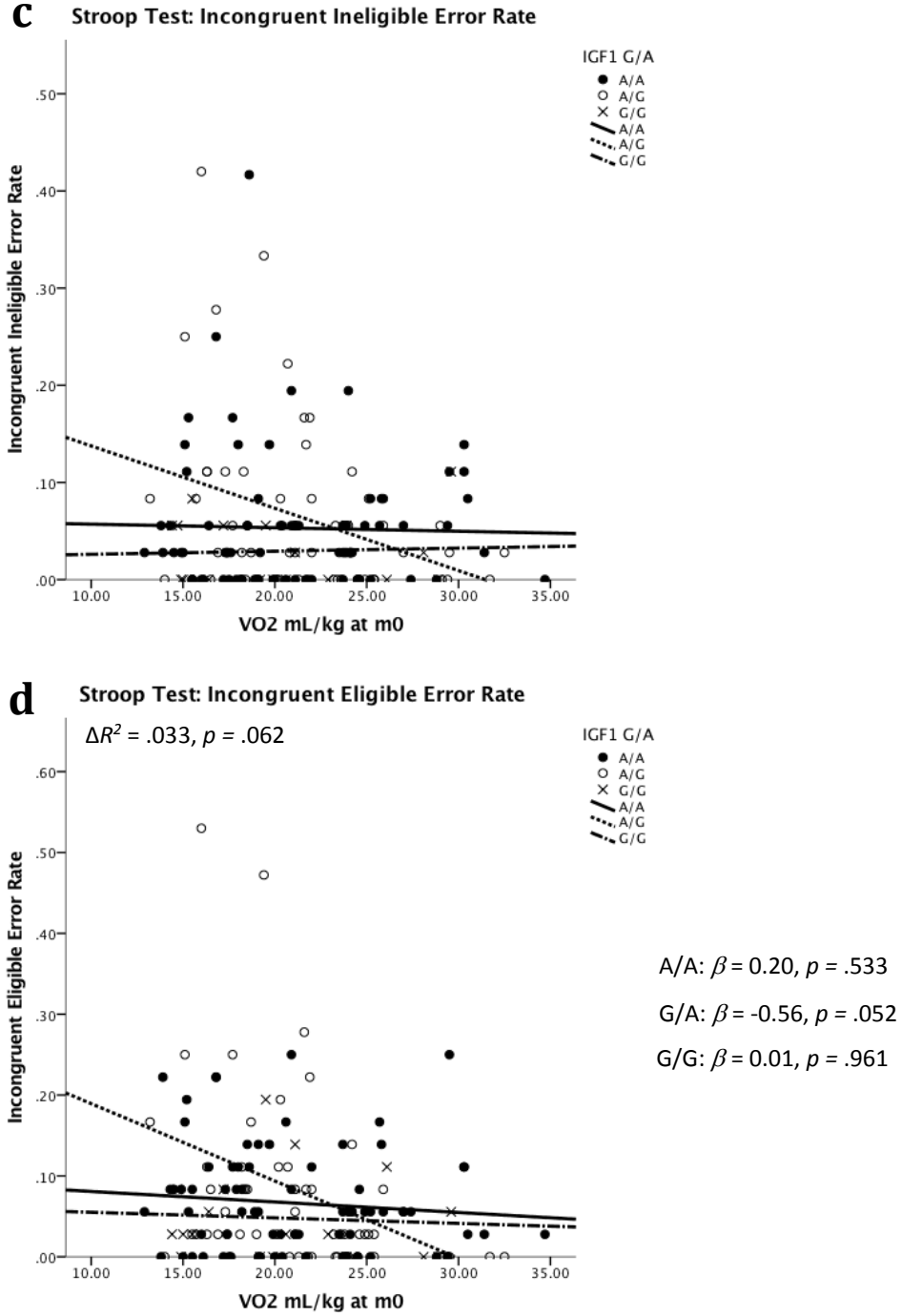
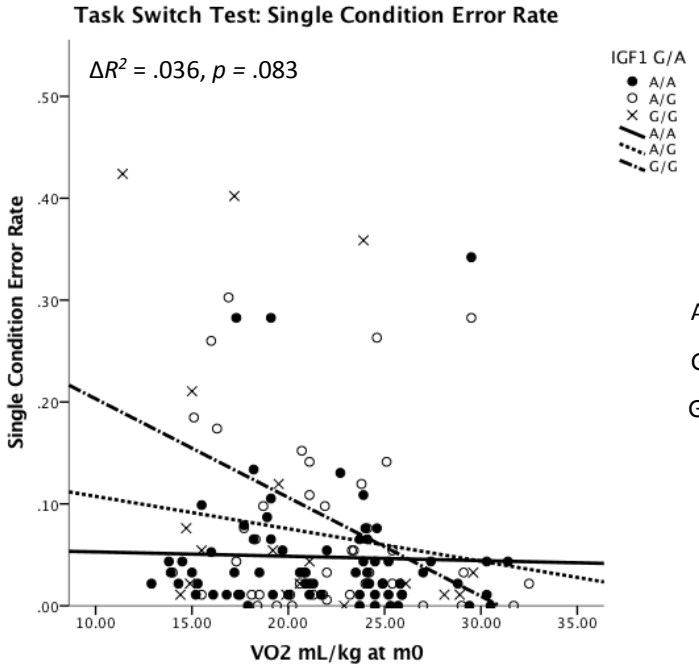


Figure 4.3 cont. Interaction effect between the IGF1 G/A SNP and cardiorespiratory fitness level (i.e. VO_2) on error rate for all trial types of the Stroop test. The interaction between IGF1 G/A x Fitness Level marginally explained variance in error rate only for the incongruent eligible trials but the trend was similar for other trial types **c)** incongruent-ineligible RT, $\Delta R^2 = 0.02$, $\Delta F(2, 142) = 1.62$, $p = .201$ **d)** incongruent eligible RT, $\Delta R^2 = 0.03$, $\Delta F(2, 142) = 2.83$, $p = .062$

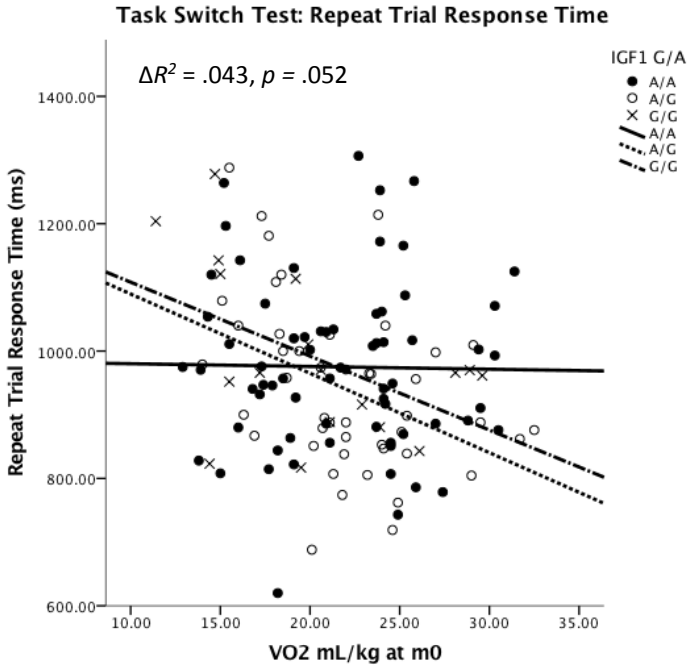


A/A: β = 0.29, p = .350

G/A: β = -0.63, p = .156

G/G: β = 0.99, p = .049

Figure 4.4 Interaction effect between the IGF1 G/A SNP and cardiorespiratory fitness level (i.e. VO_2) on error rate for the singe condition of the task switch test. The interaction marginally explained 3.6% of the variance in ER for the single condition, ΔR^2 = .036, $\Delta F(2, 122)$ = 2.54, p = .083. Simple slopes: A/A group, B = -.002, β = -0.29, $t(122)$ = -0.87, p = .350, 95% CI [-.007, .002]; G/A, B = -.005, β = -0.63, $t(122)$ = -1.61, p = .156, 95% CI [-.012, .003]; G/G group, B = -.012, β = -0.99, $t(122)$ = -2.94, p = .049, 95% CI [-.023, .000].



A/A: β = .149, p = .673

G/A: β = -.607, p = .041

G/G: β = -.288, p = .199

Figure 4.5 Interaction effect between the IGF1 G/A SNP and cardiorespiratory fitness level (i.e. VO_2) on response time for the mix condition repeat trial type of the task switch test. The interaction marginally explained 4.3% of the variance in RT for repeat trials, ΔR^2 = .043, $\Delta F(2, 122)$ = 3.02, p = .052. Simple slopes:

A/A, B = 2.65, β = .149, t(122) = 0.49, p = .673, 95% CI [-10.19, 14.39]; G/A group, B = -11.31, β = -.607, t(122) = -1.70, p = .041, 95% CI [-22.95, -1.07], G/G group, B = -8.49, β = -.288, t(122) = -0.92, p = .199, 95% CI [-26.64, 9.66]

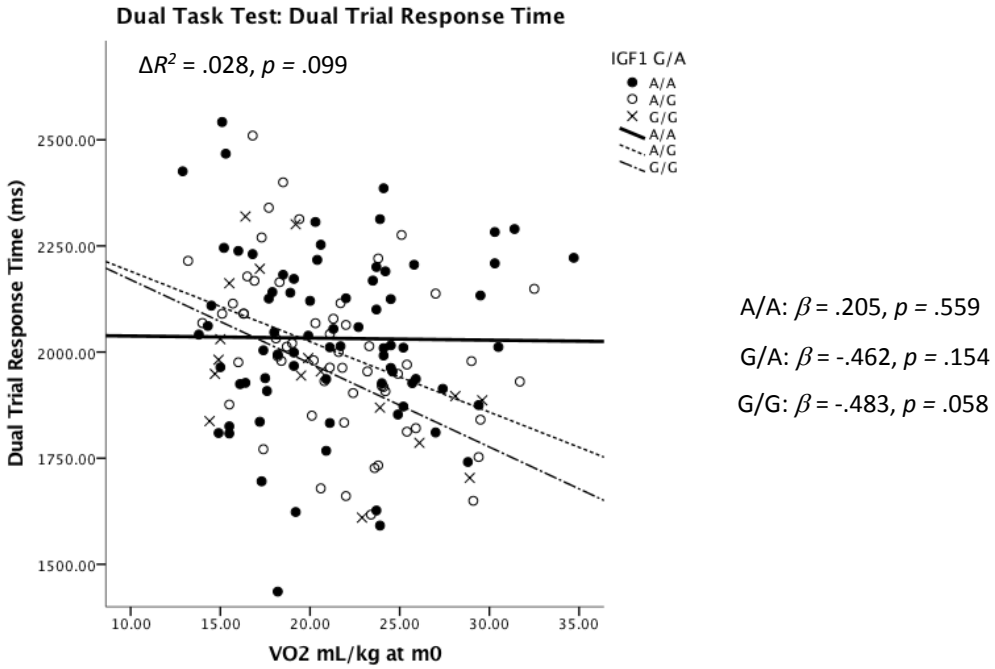


Figure 4.6 Interaction effect between the IGF1 G/A SNP and cardiorespiratory fitness level (i.e. VO_2) on response time for the dual trial type of the dual task test. The interaction marginally explained 2.8% of the variance in RT for dual trials, ΔR^2 = .028, ΔF(2, 141) = 2.35, p = .099. Simple slopes: A/A, B = 3.60, β = 0.21, t(141) = 0.70, p = .559, 95% CI [-8.50, 15.60]; G/A, B = -8.48, β = -0.46, t(141) = -1.33, p = .154, 95% CI [-21.14, 2.62]; G/G group, B = -14.26, β = -0.48, t(141) = -1.52, p = .058, 95% CI [-30.98, 0.27]

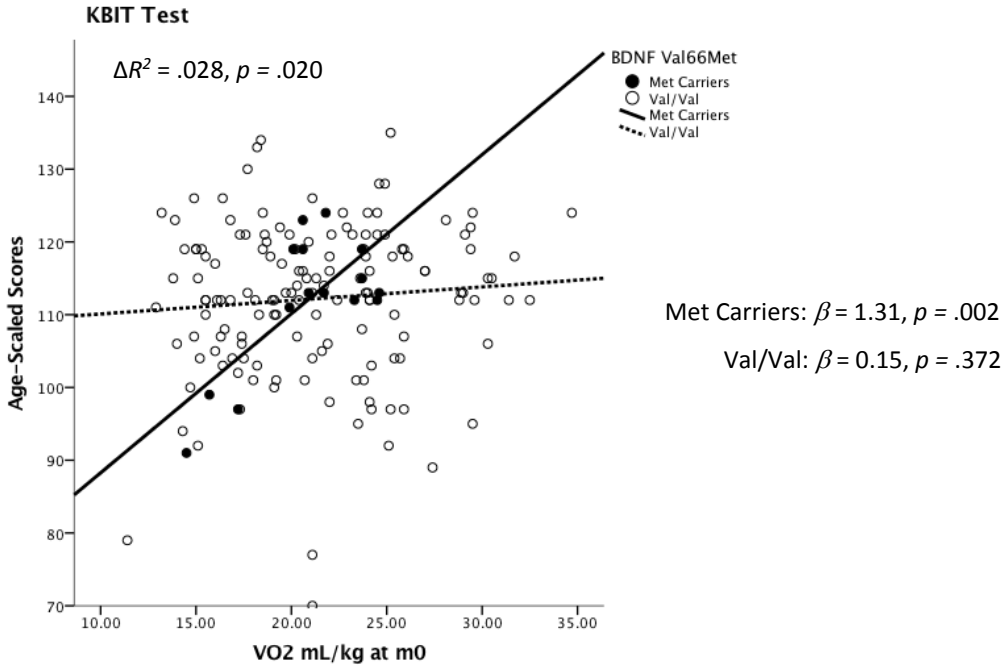


Figure 4.7 Interaction effect between BDNF Val66Met and cardiorespiratory fitness level (i.e. VO_2) on the KBIT test. The interaction explained 2.8% of the variance in age-scaled scores, $\Delta R^2 = .028$, $\Delta F(1, 152) = 5.56$, $p = .020$. Simple slopes: Met Carriers, $B = 2.22$, $\beta = 1.31$, $t(152) = 2.64$, $p = .002$, 95% CI [-0.10, 3.31]; Val/Val Homozygotes, $B = 0.20$, $\beta = 0.15$, $t(152) = 0.94$, $p = .372$, 95% CI [-0.23, 0.64]

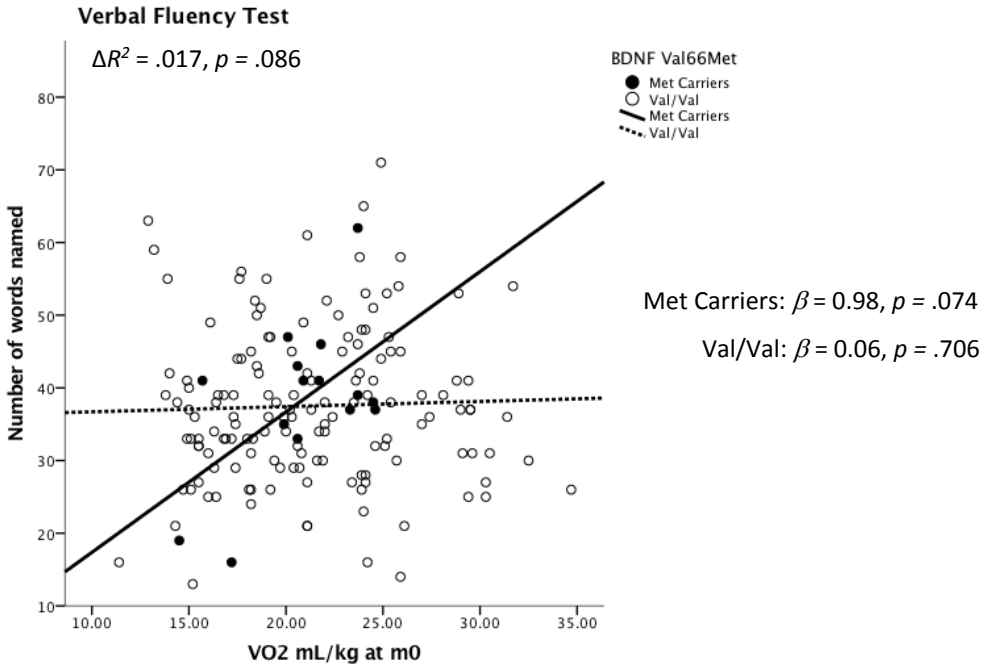


Figure 4.8 Interaction effect between BDNF Val66Met and cardiorespiratory fitness level (i.e. VO_2) on the verbal fluency test. The interaction marginally explained 1.7% of the variance in the number of words named, $\Delta R^2 = .017$, $\Delta F(1, 154) = 2.98$, $p = .086$. Simple slopes: Met Carriers, $B = 1.70$, $\beta = 0.98$, $t(154) = 1.84$, $p = .074$, 95% CI [-0.70, 3.67]; Val/Val Homozygotes, $B = 0.08$, $\beta = 0.06$, $t(154) = 0.34$, $p = .706$, 95% CI [-0.32, 0.53].

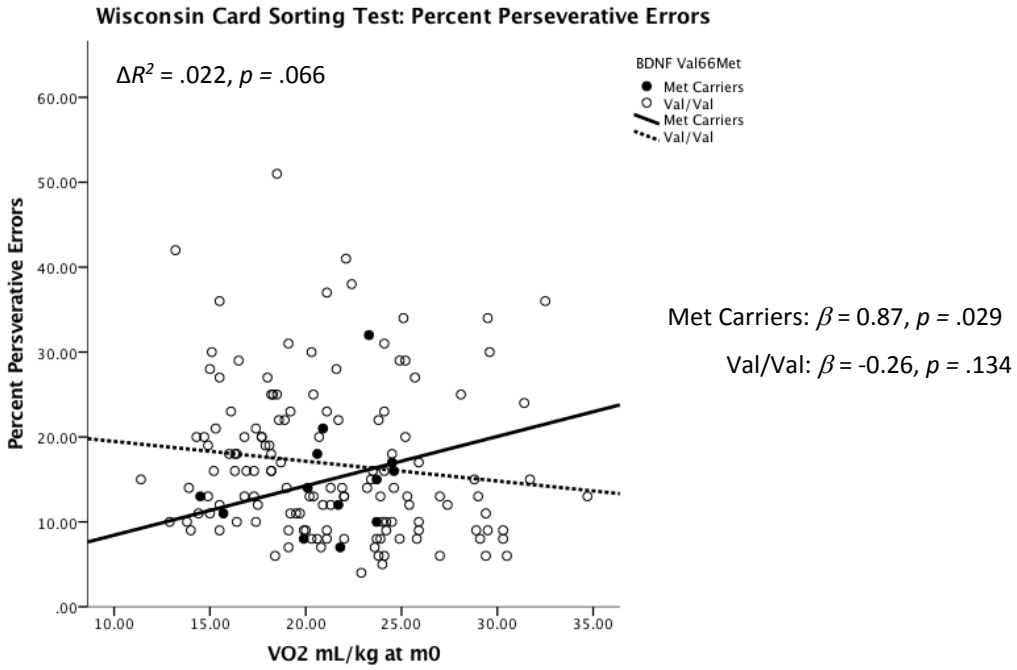


Figure 4.9 Interaction effect between BDNF Val66Met and cardiorespiratory fitness level (i.e. VO_2) on performance of the Wisconsin Card Sorting Test measured by percent perseverative errors. The interaction marginally explained 2.2% of the variance in percent perseverative errors, $\Delta R^2 = .022$, $\Delta F(1, 143) = 3.44$, $p = .066$. Simple slopes: Met Carriers, $B = 1.26$, $\beta = 0.87$, $t(143) = 1.52$, $p = .029$, 95% CI [0.20, 3.08]; Val/Val Homozygotes, $B = -0.30$, $\beta = -0.26$, $t(143) = -1.55$, $p = .134$, 95% CI [-0.71, 0.07].

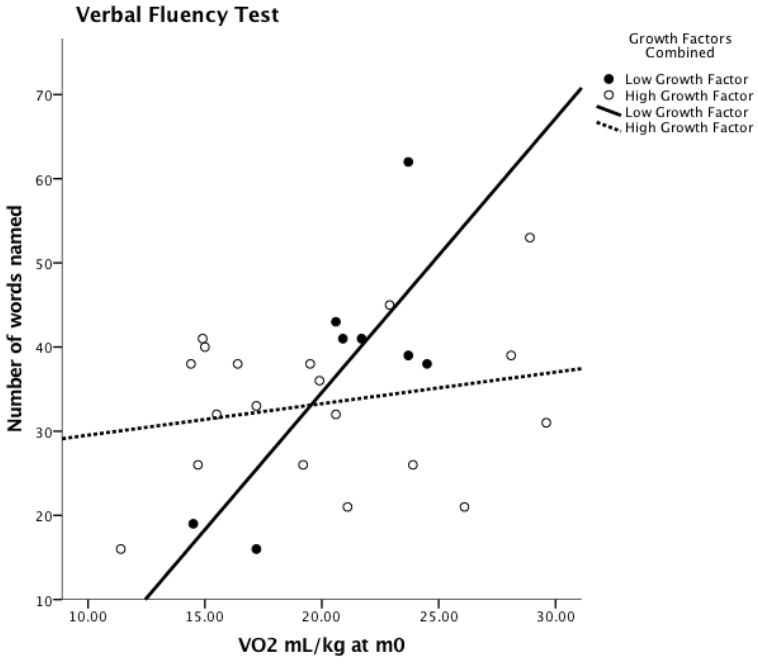


Figure 4.10 Interaction effect between the growth factor (GF) genes combined and cardiorespiratory fitness level (i.e. VO_2) on performance of the verbal fluency test. The interaction significantly explained 11.3% of the

variance, ΔR^2 = .113, $\Delta F(1, 20) = 5.74$, p = .026. Simple slopes: low GF group, B = 2.92, β = 2.65, $t(20) = 3.09$, p = .014, 95% CI [-0.80, 5.86]; high GF group, B = 0.55, β = 0.50, $t(20) = 1.04$, p = .408, 95% CI [-0.48, 1.78].

Chapter 5: General Discussion

This study was conducted as an exploratory investigation into the interactive effects of genes and cardiorespiratory fitness level on cognition in healthy older adults. Specific genes were chosen because of the neurobiological significance of the protein products. Specifically, two dopamine related genes, Catechol-O-Methyltransferase (COMT) and Dopamine Beta Hydroxylase (DBH), each with two different single nucleotide polymorphisms (SNPs); and two growth factor related genes, Brain-derived Neurotrophic Factor (BDNF) and Insulin-like Growth Factor I (IGF1). In animal models it is possible to measure and manipulate neurochemical levels (e.g. gene knock outs, microdialysis, or direct injection of a neurochemical or receptor agonist/antagonist into the brain) and observe its effects on behavior. However, this level of control or even monitoring of any neurochemical level within the human brain can be difficult and impractical. Enter natural genetic variation. By investigating naturally occurring variation in genes whose products have well-defined biological actions, it is possible to infer high-medium-low neurochemical groups (Raz & Lustig, 2014) and observe their effects on behavior.

Further, considering external factors and investigating potential interactions with genetics acknowledges the complexity of what contributes to variability in behavior. Twin studies lay the foundation for the heritability of cognitive abilities and the increasing influence of genetics on various aspects of cognition in later life. Therefore, genetic and interactive effects not seen in young and middle adult populations may be elucidated through older adult populations. Studying the effects of genetics and its interaction with positive lifestyle factors, such as exercise or physical activity, is also interesting as it could lead to easy ways to counterbalance negative genetic predisposition. For example, there is already a company offering personalized weight loss and fitness plans based on genetic status (www.fitnessgenes.com), and it is conceivable that the future holds individualized cognitive therapies based on genetic status.

Dopamine Related Genes

Dopamine (DA) related genes were chosen due to DA's known role in cognitive functioning (Cools & Robbins, 2004; Seamans & Yang, 2004), as well the contribution

aging-related decline in the DA system has toward aging-related cognitive decline (Cabeza, 2001a). Dopamine Beta Hydroxylase (DBH) and Catechol-O-Methyltransferase (COMT) were chosen because of their influence on the variability of dopamine (DA) levels in the body and brain. I investigated the genotypic effect of each SNP for each gene independently and then brought cardiorespiratory fitness into the mix. There are two perspectives here: 1) can a "bad" genotype be rescued? and 2) can genetics help explain the variability in the relationship between cardiorespiratory fitness level and cognition? Based on aging and exercise effects on the DA system, I predicted cardiorespiratory fitness level would have a positive influence on performance for tests of memory and executive control. I expected this relationship to be strongest for those with the allele that leads to the lowest DA levels under the assumption they had the most to gain.

Dopamine Beta Hydroxylase

Dopamine Beta Hydroxylase (DBH) is an enzyme that converts dopamine (DA) into norepinephrine (NE) and variation in the DBH gene leads to variability in the DA:NE ratios in the brain (Cubells & Zabetian, 2004). I investigated two common single nucleotide polymorphisms (SNP), DBH 444 G/A and DBH -1021 C/T, related to the DBH gene. Previous literature showed the groups with alleles translating to higher DBH levels and thus lower DA:NE ratios (i.e. G allele for the DBH444 G/A SNP and C allele for the DBH -1021 C/T SNP) showed better cognitive functioning. Specifically, spatial memory performance increased with increasing G allele load for the DBH 444 G/A SNP (Greenwood et al., 2009; Parasuraman et al., 2005), and better response inhibition was seen with increasing C allele load for the DBH -1021 C/T SNP (Greene et al., 2009). However, these studies used middle aged or a mixed middle and older adult samples. I predicted in our sample of older adults, we would observe a reversal of this genetic effect such that the groups with the allele that is believed to translate to lower DBH levels and in turn higher DA:NE (i.e. A allele for DBH 444 G/A and T allele for the DBH -1201 C/T) would show better performance on various tests of memory and executive control and that is what I found.

For the DBH 444 G/A analysis, results point to the A allele leading to better maintenance and switching of sets. For the DBH -1021 C/T SNP, T Carriers showed better

performance on a test of working memory and were faster on a test of spatial memory. Furthermore, T Carriers had better performance on a test of verbal fluency and were found to be faster across several tasks of executive control. In all cases, there were no differences found between the groups on error, implying there was no sacrifice of accuracy for speed of response. These results are consistent with a more recent study that found cognitive costs to the C allele of the DBH -1021 C/T SNP only in their older adult group (Greenwood, Lin, Sundararajan, Fryxell, & Parasuraman, 2014).

DBH genotypic effects. The DBH 444 G/A SNP showed a genotypic effect on executive function, specifically maintenance and coordination of multiple task goals. This effect was seen on error rates (ER) of the task switch test. For the single condition, the G/A group showed the lowest error rates, while the G/G group showed the highest error rates during the mix condition (Figure A.1a). Overall, the A/A group showed the lowest global error cost (i.e. mix condition ER - single condition ER; Figure A.1b). Further, the G/G group had the highest errors on both the repeat and single trials of the mix condition (Figure A.2). Taken together, the results support the prediction that the A allele of the DBH 444 G/A SNP leads to better executive functioning.

The DBH -1021 C/T SNP had effects on both memory and executive control tests. Specifically, T Carriers were faster across all memory loads of the spatial memory test while showing no differences on error rate (Figure A.3). T Carriers were also faster on both trial types of the Flanker test, with no group differences on error rate for either trial type, suggesting T Carriers were generally faster and had better inhibitory processing compared to C/C Homozygotes. Further, T Carriers showed better maintenance and coordination of multiple task goals, evidenced by better performance on both the task switch test and the dual task test. For the task switch test, T Carriers had faster response times on the more difficult mix condition of the task switch test (Figure A.5a) and lower global cost scores (i.e. mix condition – single condition) for both response time and error rate (Figure A.5b and d respectively). Additionally, within the mix condition, T Carriers were significantly faster on switch trials and marginally faster on repeat trials (Figure A.6), while showing no group differences on error rate for either trial type. For the dual task test, T Carriers were significantly faster on single trials and marginally faster on dual trials (Figure A.7), with no

group differences on error rate. These results suggest that carriers of the T allele for the DBH -1021 SNP are more effective at both shifting and time-sharing.

Further, there was an additive effect of the two DBH SNPs on executive functioning. The DBH High DA group showed better inhibitory processing through significantly faster response times on both congruent and incongruent trials of the Flanker test (Figure A.8). This group also showed a trend toward more effective maintenance and coordination of multiple tasks through marginally lower global error cost (i.e. mix condition ER – single condition ER; Figure A.9).

Collectively, these results follow the prediction that possession of the alleles that translate to higher DA levels (i.e. A allele for DBH 444 G/A and T allele for DBH -1021 C/T) would lead to better cognitive functioning.

DBH x Fitness Level interactive effects. Results also showed DBH genotype moderated the effect of cardiorespiratory fitness level (CRF; measured by VO_2) on cognitive test performance in this sample of healthy sedentary older adults. Specifically, the DBH 444 G/A SNP moderated the effect of CRF on maintenance of multiple task goals and shifting. The G/G and A/A groups both showed a significant decrease in errors during the mix condition with increasing VO_2 scores (Figure 3.1b). This effect was driven by the G/G and A/A groups showing a decrease in errors during switch trials with increasing VO_2 (effect was marginal in A/A group; Figure 3.2b).

For the DBH -1021 C/T SNP, there was a significant interaction effect on working memory measured by the backward span test, suggesting the relationship between CRF and working memory differs for T Carriers versus C/C Homozygotes (Figure 2.3b). Although simple slopes analysis did not reveal a significant relationship between VO_2 and backward span length, the trend was a positive relationship in T Carriers. Further, the DBH -1021 C/T SNP moderated the effect of CRF on cognitive flexibility measured by total errors on the Wisconsin Card Sorting Test. Again, only T Carriers showed a decrease in total errors with increasing VO_2 (Figure 3.4).

The analysis with the combined DBH SNPs showed an interactive effect on shifting as measured by the task switch test. The DBH Low DA group showed a significant decrease in local ER cost (i.e. switch trial ER – repeat trial ER) with increasing VO_2.

My prediction was that the group with the alleles translating to lower DA levels (i.e. G allele for DBH 444 G/A and C allele for DBH -1021 C/T) would show the greatest benefit from increasing CRF. Although, the combined DBH SNPs followed my prediction, a clear pattern did not emerge from the results of the individual SNPS, suggesting each SNP may moderate the relationship differently. This study suggests there is an interactive effect of the DBH gene and cardiorespiratory fitness level on cognition in healthy older adults, but further investigations with greater power are needed to elucidate the nature of the interaction.

Catechol-O-Methyltransferase

The Catechol-O-Methyltransferase (COMT) gene encodes for a methylation enzyme that metabolizes catecholamines including dopamine (Axelrod & Tomchick, 1958), and plays a key role in the prefrontal cortex (PFC) where there is a low level of dopamine transporters (DAT; Chen et al., 2004). COMT was chosen as a candidate gene because of its potential to control the basal DA neurotransmission levels within the PFC, which could contribute to individual differences in human behavior. Two common SNPs in the COMT gene were chosen for investigation: 1) COMT Val158Met, which effects enzymatic activity, and 2) COMT C/G, which affects enzyme availability.

COMT Val158Met is a functional SNP that causes a valine (Val) to methionine (Met) switch in the amino acid sequence at codon 158. This change to Met produces a less thermostable enzyme, leading to a 40% decrease in enzymatic activity and resulting in higher PFC DA levels (Chen et al., 2004; Lotta et al., 1995; Savitz et al., 2006). The COMT C/G SNP affects mRNA secondary structure, consequently affecting protein synthesis, but not the functioning of the enzyme itself (Nackley et al., 2006). Carriers of the C allele are believed to have lower COMT activity due to less efficient protein synthesis (Diatchenko et al., 2005; Nackley et al., 2006).

The results from this study also add to the many findings related to the COMT Val158Met SNP and extend the findings of the less investigated SNP, COMT C/G. Further, the study explores the results of an additive effect of the two polymorphisms on cognition as well as the interactive effect of the COMT gene with cardiorespiratory fitness level in a sample of healthy older adults.

COMT genotypic effects. The COMT Val158Met SNP had an effect on both memory and executive control tests, with Val/Val homozygotes generally showing better performance. Specifically, Val/Val homozygotes showed better working memory as evidenced by marginally larger backward span lengths compared to both Val/Met heterozygotes and Met/Met homozygotes (Figure A.10). Val/Val homozygotes were also significantly faster on all memory loads of the spatial memory test (Figure A.11), with no differences found on error rate.

On tests of executive control, Val/Val homozygotes were significantly faster on both congruent and incongruent trial types for the Flanker test (Figure A.12a), but this group also showed the highest error rates on incongruent trials (Figure A.12c) and ER cost (i.e. incongruent trial ER – congruent trial ER; Figure A.12d). There is potential that the Val/Val group sacrificed accuracy for speed, therefore I am hesitant to interpret these results since the instructions to favor accuracy over speed were not followed.

However, the COMT Val158Met SNP did have an effect on coordination, specifically time-sharing, with the Val/Val group showing the fastest response times on both single and dual trials of the dual task test (Figure A.13a and b), and a marginally lower ER cost (i.e. dual trial ER – single trial ER; Figure A.13d).

The COMT C/G SNP also had an effect on both memory and executive control tests. Specifically, G Carriers showed marginally larger backward span lengths (Figure A.14) and were faster on all memory loads of the spatial memory test with no differences found on error rate (Figure A.15). These results suggest that the COMT C/G SNP influences working and spatial memory. Furthermore, G Carriers showed marginally better inhibitory processing as evidenced by faster RTs on incongruent trials of the Flanker test (Figure A.16), with no differences on error rate. G Carriers also showed a significantly lower RT cost (i.e. dual trial RT – single trial RT; Figure A.17b) compared to C/C Homozygotes, suggesting they were better at time-sharing.

When both COMT SNPs were combined, there was an additive effect on the tests of working and spatial memory. The COMT Low DA group (i.e. Val/Val + G Carriers) had significantly longer backward span lengths and were faster across all memory loads of the spatial memory test. Further, the COMT Low DA group showed better time-sharing ability.

They were significantly faster on both single and dual trials of the dual task test and showed a significantly lower RT cost to the dual trial type.

Collectively, the results are in the opposite direction of prediction. Based on the extant literature and the assumption that the Val allele of the COMT Val158Met SNP and the G allele of the COMT C/G SNP lead to lower prefrontal dopamine availability, I expected Val/Val homozygotes and G Carriers to show worse performance on the tests of memory and executive control functioning. These results add to the equivocal literature and implicate the need to take other factors into consideration such as gene x gene or gene x external factor interactions.

COMT x Fitness Level interactive effects. When cardiorespiratory fitness level (CRF) was taken into account, I did find that the COMT Val158Met SNP moderated its effect on working memory such that only the Val/Val group showed an increase in backward span length with increasing VO_2 (Figure 3.6b). There was also a marginal interactive effect on processing speed measured by congruent trials of the Flanker test. Results suggest the relationship between CRF and response time in congruent trials are different, but there was not enough power to tease apart the different directions for each group (Figure 3.7). On the dual task test, somewhat oddly, Val/Met heterozygotes showed an increase in the RT cost (i.e. dual trial RT – single trial RT; Figure 3.8) with increasing cardiorespiratory fitness level. This was unexpected since increases in cardiorespiratory level are typically associated with increases in performance on a variety of cognitive tests.

The COMT C/G SNP moderated the relationship between CRF and executive functioning. Specifically, there was a significant interaction on the WCST with the C/C Homozygotes showing a marginal decrease in percent perseverative errors with increasing VO_2. There was also an interactive effect on the dual task test, with G Carriers showing a significant increase in RT cost (i.e. dual trial RT – single trial RT), which again was unexpected for the same reason as mentioned above.

When the two COMT SNPs were combined, there was an interactive effect on working memory with the COMT Low DA group showing an increase in backward span length with increasing VO_2. There was also a marginal effect on the WCST with the COMT High DA group showing a decrease in percent perseverative errors with increasing VO_2.

My prediction was that the alleles translating to lower prefrontal dopamine (i.e. Val allele on COMT Val158Met and G allele on COMT C/G) would benefit the most from an increased CRF, but the results of this study are difficult to interpret. Taken together, they suggest the COMT gene may moderate the relationship between cardiorespiratory fitness levels on cognition in healthy older adults, but further investigation is needed to discern the pattern of this moderation.

Overall, these results imply that carriers of an allele that leads to higher COMT activity and thus lower DA levels show better performance on tests of spatial memory and executive functioning, specifically inhibition and multi-tasking. There is a hypothesis that Met Carriers of the COMT G/A SNP have higher PFC dopamine levels, allowing DA to diffuse away from the activated synapse and activate extrasynaptic D1 receptors. This D1 activation enhances input-related neuronal excitability and increases inhibitory feedback to neurons not receiving input. On the other hand, in Val carriers who are thought to have less PFC DA levels, the DA remains local to the activated synapse and activates D2 receptors, which reduces inhibitory interneuron activity, which leads to increases in non-specific neuronal excitability. So Met carriers are used to "working" with a clearer signal to noise ratio, and with age-related declines in DA levels and D1 receptors, the shift from an efficient to a noisy system is detrimental. In other words, the age-related decline in D1 receptors could be more detrimental to Met carriers because of the added noise to the signal (Tunbridge, Harrison, & Weinberger, 2006).

However, since Val carriers are used to "working" with a noisy system, the age-related declines in DA levels and D2 receptors may not have as detrimental an effect on them as for Met carriers. In fact, D2 receptor decline may actually help, because the inhibitory interneurons would not be activated by local DA, and with increasing task demands, the increased DA may be helpful. This same reasoning can be extended to the results from the COMT C/G SNP.

Further, when cardiorespiratory fitness level is taken into account, there may be differential results due to the differences in the degree to which each test taxes the DA system. That is to say, the effects that cardiorespiratory fitness level has on the DA system (e.g. increased DA receptor density) may lead to benefits in one test, but be detrimental in another test for particular genotypes.

Growth Factor Genes

Insulin-like Growth Factor I

Insulin-like growth factor I (IGF1) belongs to a family of growth factors known as somatomedins that play an important role in mammalian growth and development (Froesch et al., 1985). IGF1 contributes to brain development and overall brain health, evidenced by its diffuse expression pattern including the cortex, hippocampus, cerebellum, brainstem, hypothalamus and the spinal cord (Bach 1991). IGF1 has also been found to support cognitive ability (Arwert et al., 2005; Trejo et al., 2008; van Dam & Aleman, 2004). Further, exercise has been shown to increase IGF1 levels and blockage of IGF1 receptors eliminates exercise-induced augmentation of recall in rats (Ding et al., 2006).

The IGF1 gene was selected for investigation because 40-60% of variation in circulating IGF1 levels is determined by genetic factors (Harrela et al 1996; Verhaeghe et al 1996; Hong et al 1997; Hall et al 1999), and there is a common SNP resulting in a G to A switch in exon 4 of the 3' untranslated region containing regulatory domains for mRNA expression (rs6220; http://www.ncbi.nlm.nih.gov/SNP/snp_ref.cgi?rs=rs6220). This SNP may alter the RNA stability and regulation of protein expression (Al-Zahrani et al., 2006; Huuskonen et al., 2011), and studies have shown that this SNP is associated with circulating levels of IGF1 protein such that the minor G allele is associated with higher levels of circulating IGF1 (Diorio et al., 2008; Johansson et al., 2007; Verheus et al., 2008).

To date**,** there are no studies investigating the relationship between the IGF1 G/A SNP and cognition. However, in a population-based study (N = 278), Licht et al., (2014) found no association between a different IGF1 polymorphism (i.e. variable length cytosine-adenine repeat sequence) and cognitive functioning in their middle aged sample. Therefore, the results of this study are novel and predictions were based on the relationship between the IGF1 G/A SNP and IGF1 serum levels, and the effect of IGF1 serum levels on cognitive functioning. Since the IGF1 G allele has been associated with higher levels of circulating IGF1 (Diorio et al., 2008; Johansson et al., 2007; Verheus et al., 2008), and in general, there is a positive association between IGF1 levels and cognitive functioning (Arwert et al., 2005; Bellar et al., 2011), I expected G carriers to perform better on both

tests of memory and executive functioning. Further, due to aging-related decreases in IGF1 levels and exercise-induced increases, I expected A/A homozygotes to show cognitive benefits from higher cardiorespiratory levels.

IGF1 genotypic effects. Following prediction, I found a genetic effect of the IGF G/A SNP on spatial memory. The G/A group was faster across all memory loads, and did not show differences on error rate. The IGF1 G/A polymorphism also showed effects on a few tests of executive control, and mostly followed my predictions. On the Stroop test, the G/G group was significantly faster than the other two groups on incongruent-ineligible trials. Although there were no significant differences for the other trial types, the trend remained the same. Further, on the dual task test, the G/G group showed the lowest RT cost, followed by the G/A, then A/A groups. The G/G group also showed the lowest error rate on both single and dual trial types. For the task switch test, the A/A group showed marginally less errors on switch trials compared to the G/A and G/G (highest errors) groups. This departure from prediction could point IGF1 having a differential effect on time-sharing versus shifting, but further investigation with more power would be required to confirm and elucidate this potential difference.

Taken together, if the inference that the G allele leads to higher IGF1 levels holds true, these results are mostly consistent with the literature in which higher circulating IGF1 levels were associated with better performance in a variety of cognitive domains including memory and executive functioning (Aleman et al., 1999; Rollero et al., 1998; van Dam & Aleman, 2004; Vitiello et al., 2006).

IGF x Fitness Level interactive effects. When cardiorespiratory fitness level was taken into consideration, the interaction between IGF1 G/A x Fitness Level had an effect on spatial memory. The G/A group showed a significant decrease in errors with increasing VO_2 for Memory Load 2, implying that moderate levels of IGF1 lead to better spatial memory.

For the Stroop test, increasing levels of VO_2 led to a significant decrease in response time across all trial types and a decrease in error rate for incongruent-ineligible trials for the G/A group. Again, this trend was seen across all trials. These results suggest higher levels of IGF1 may benefit response inhibition, and increased levels of cardiorespiratory fitness can benefit those expected to have moderate levels of IGF1 (i.e. G/A heterozygotes).

On the task switch test, the G/G group showed a significant decrease in errors during the single condition with increasing VO_2 levels. The G/A group showed a significant decrease in RT for repeat trials with increasing VO_2 levels. For the dual task test, the G/G group showed a marginal decrease in RT with increasing VO_2 during dual trials. These results suggest higher IGF1 levels are good for multiple set coordination.

Overall, the results imply those with a genetic predisposition for moderate to higher levels of IGF1 benefit the most from increased cardiorespiratory fitness level.

Conclusions. The present study showed for the first time that the IGF1 G/A genotype (rs6220) may influence cognitive performance in healthy sedentary older adults, specifically spatial memory and executive functioning.

Results imply that having a G Allele may lead to better spatial memory, inhibitory processing and multi-tasking. Further, cardiorespiratory fitness level seems to have a positive association with cognitive functioning in G allele carriers, suggesting that cardiorespiratory fitness may benefit those that are genetically predisposed to higher levels of IGF1. That is to say, since IGF1 levels decrease with increased age, having the G allele may provide some cognitive protection, and higher levels of cardiorespiratory fitness can help even more.

It is difficult to determine what is truly driving these effects, since variance in cardiorespiratory fitness level alone may not be enough to have an effect on cognitive performance. For example, in mice that were showing impaired spatial learning and reduced hippocampal adult neurogenesis due to low serum IGF1 levels, running exercise alone was not able to restore spatial learning ability or hippocampal neurogenesis – restoration only occurred with external IGF1 administration (Trejo et al., 2008).

The IGF1 G/A genotype may have a real effect on cognitive performance in healthy older adults, and future investigations with larger sample sizes and consistent sampling of serum IGF1 levels would strengthen and further elucidate this relationship. Future investigations could not only expand the understanding of the association between IGF1 G/A genotype and serum levels, but also the effects of serum levels on cognitive performance.

Brain-derived Neurotrophic Factor

Brain-derived neurotrophic factor (BDNF) belongs to a family of growth factors called neurotrophins which are regulatory factors that mediate the differentiation, proliferation and survival of cholinergic, dopaminergic and serotonergic neurons (Savitz et al., 2006). Although BDNF is expressed throughout the brain, its effects are most apparent in the prefrontal cortex (PFC) and hippocampus (HC; Savitz et al., 2006). A common functional SNP in the BDNF gene (rs6265), G to A switch at nucleotide 196, results in a Valine (Val) to Methionine (Met) amino acid substitution at codon 66 (Val66Met). This SNP is located in the 5' pro-BDNF sequence and it is unlikely that it affects the actual biological activity of the mature protein. Instead, it is believed to alter the intracellular processing and regulated activity-dependent secretion of mature BDNF (Egan et al., 2003). The Met allele is thought to selectively impair secretion and intracellular trafficking of BDNF in primary cortical neurons and neurosecretory cells (Egan et al., 2003; Savitz et al., 2006).

The extant literature suggests Met allele carriers have reduced hippocampal volume (Bueller et al., 2006; Erickson et al., 2010; Pezawas et al., 2004), and cognitive impairments (Dincheva, Glatt, & Lee, 2012; Dincheva et al., 2012; Egan et al., 2003; Hariri et al., 2003; Miyajima et al., 2008).

BDNF genotypic effects. My findings were not consistent with the literature in that Met Carriers showed a significantly lower RT cost in the dual task test. Although the allelic distribution did follow the Hardy-Weinberg equilibrium, I hesitate to draw conclusions from these results due to the small size of the Met group. Further investigations with a better distribution and more power to differentiate the effects between the groups are merited.

BDNF x Fitness Level interactive effects. In relation to cardiorespiratory fitness level, Met Carriers showed a marginal increase in the number of words named during the verbal fluency test with an increase in VO_2. And, oddly, the Met carriers also showed an increase in percent perseverative errors with an increase in VO_2 on the Wisconsin Card Sorting Test. Again, the small size of the Met group most likely contributes to this odd result, but these results could also suggest a more complex relationship between cardiorespiratory fitness level and cognition. That is to say, higher is not always better; and other factors, such as genetics and external risk factors (not investigated here), may play a

role in the direction of the association between cardiorespiratory fitness level and cognition in healthy older adults.

Conclusions. Results implied Met carriers of the BDNF SNP were better at multi-tasking and verbal fluency, but also that an increase in cardiorespiratory fitness level may lead to more perseverative behavior. The low sample size for the Met Carriers in this sample make it difficult to detect true differences in the present study, even though the distribution of the BDNF alleles did conform to Hardy-Weinberg expectations.

There is no doubt that BDNF is important to the development of a healthy brain, but evidence that varying levels due to a genetic basis influence higher level brain functioning is equivocal and further research with larger sample sizes is necessary.

Growth Factor (GF) Genes Combined

GF Genes genotypic effects. When the two SNPS were combined into low and high growth factor (GF) groups, the low GF group performed significantly better on backward span. This was unexpected since growth factors are thought to have a protective effect against age-related cognitive declines through increases in brain health.

GF Genes x Fitness Level interactive effects. However, the low GF group also showed a significant increase in the number of words named during the verbal fluency test with increasing VO_2 suggesting increased cardiorespiratory fitness levels is beneficial to the cognitive flexibility of those with lower levels of growth factors. This analysis was purely exploratory and interpretations should be made cautiously due to the small group sizes.

Caveats and Future Directions

This study was undertaken as an exploratory analysis. One of the main limitations of this study was the sample size. Although the sample size was large for a functional neuroimaging study and a randomized controlled trial, it was small for a genetic analysis. Further, the cross-sectional nature of this particular analysis does not allow for knowledge of what contributed to the variability in participants' cardiorespiratory fitness level. On top of that, this study was conducted on healthy older adults that were specifically recruited

because they were sedentary. This contributes to the small range of cardiorespiratory fitness in this sample.

A strength and a weakness of this study was the testing of different domains of cognitive behavior with a wide variety of tests. The strength is in the careful choosing of tasks to test associations with specific cognitive behaviors. And considering this was an exploratory investigation, analysis of individual performance measures is a good place to start. However, results may be more generalizable if performance measures were investigated: 1) where applicable, within task measures were standardized and combined to take both speed and accuracy into account or 2) latent factors created based on theoretical or statistical similarity. An investigation into intra-individual variability of performance measures would also be of interest to see if it is more sensitive to genetic variation in a healthy older adult population. Further, a longitudinal as opposed to cross-sectional analysis could reveal what role genetics has on the trajectory of cognitive aging, and how external factors may speed up or slow down aging related deficits (i.e. brain structure, function, or cognition).

The future of the analysis of genetic influence on cognitive ability is moving away from single candidate gene analyses since each has such a small effect, and each gene does not act alone. It is important to take into consideration status of other gene variants and external factors such as risk factors for disease and positive lifestyle factors like exercise and physical activity (Raz & Lustig, 2014). Other alternative methods include genome wide associates studies (GWAS), proteomics and epigenetics. GWAS employs whole-genome sequencing to detect associations between rare variants and complex traits and is hypothesis-free. This allows for investigations of up to 1 million SNPs at once, to identify genomic regions associated with particular traits or common genetic variation associated with diseases. Technological advancement has made it affordable and feasible to sequence the exomes of large numbers of individuals, allowing sufficient power to detect these associations (for review Harris & Deary, 2011). In proteomics, one can look directly at the proteins under the control of multiple genetic variants by using methods such as mass spectrometry. This technique can help in the understanding of the biological foundations of cognitive differences.

Epigenetic mechanisms refer to DNA modifications that do not alter the sequence of the DNA, but change gene expression and in turn cell function (Harris & Deary, 2011; Mather, Kwok, Armstrong, & Sachdev, 2014). Examples of epigenetic modifications and mechanisms include post-translational histone modification, DNA methylation, and non-coding RNAs. The investigation of epigenetics in cognitive aging is in its early stages, but there have been both animal and human studies that have observed epigenetic changes with cognitive aging (Mather et al., 2014; Spiegel, Sewal, & Rapp, 2014).

In conclusion, this thesis study has shown that a relationship between cognition and genetic variation in genes related to specific neurochemicals may be modified by cardiorespiratory fitness level in a sample of healthy sedentary older adults. Further, it highlights the importance of exploratory analyses to help guide future interests.

Appendix A: Dopamine Related Genes Supplemental Tables and Figures

Tables for DBH 444 G/A genotypic analysis

Table A.1 Summary of ANCOVA analysis of the effect of DBH 444 G/A SNP on performance of the spatial memory test

	F (1, 170)	*p*
Memory Load 1		
Response Time	1.200	.304
Error Rate	0.298	.743
Memory Load 2		
Response Time	0.954	.387
Error Rate	0.418	.659
Memory Load 3		
Response Time	0.730	.483
Error Rate	0.255	.775

Tables for DBH Combined analysis

Table A.2 Summary of ANCOVA analysis of the effect of DBH Combined on performance of the task switch test

	F (1, 36)	*p*
Single Condition		
Response Time	0.507	.481
Error Rate	1.244	.272
Mix Condition		
Response Time	1.610	.213
Error Rate	0.423	.520
Repeat Trial Type		
Response Time	1.933	.173
Error Rate	0.170	.683
Switch Trial Type		
Response Time	0.931	.341
Error Rate	0.645	.427

Tables for COMT Val158Met genotypic analysis

Table A.3 Summary of boostrapped pairwise comparisons for the effect of COMT Val158Met on response time for the spatial memory test

			95% Confidence Interval	
Memory Load 1	Mean Difference	*p*	Lower	Upper
Val/Val v Val/Met	-73.730*	.037	-139.692	-3.123
Val/Val v Met/Met	-122.100*	.002	-197.219	-47.997
Val/Met v Met/Met	-48.370 +	.096	-105.126	9.723

			95% Confidence Interval	
Memory Load 2	Mean Difference	*p*	Lower	Upper
Val/Val v Val/Met	-67.082 +	.058	-135.481	3.476
Val/Val v Met/Met	-111.550*	.008	-190.593	-31.449
Val/Met v Met/Met	-44.468	.134	-105.186	10.305

			95% Confidence Interval	
Memory Load 3	Mean Difference	*p*	Lower	Upper
Val/Val v Val/Met	-66.363 +	.082	-142.641	7.185
Val/Val v Met/Met	-143.891*	.001	-228.446	-57.222
Val/Met v Met/Met	-77.527*	.017	-140.269	-15.371

* $p \leq .05$; + $p \leq .15$

Table A.4 Summary of boostrapped pairwise comparisons for the effect of COMT Val158Met on response time and error rate for the Flanker test

			95% Confidence Interval	
Congruent Trial RT	Mean Difference	*p*	Lower	Upper
Val/Val v Val/Met	6.095	.763	-33.392	46.638
Val/Val v Met/Met	-26.986	.242	-70.927	19.814
Val/Met v Met/Met	-33.081*	.031	-62.713	-3.843

			95% Confidence Interval	
Incongruent Trial RT	Mean Difference	*p*	Lower	Upper
Val/Val v Val/Met	9.354	.695	-37.810	55.639
Val/Val v Met/Met	-38.010	.158	-89.795	14.624
Val/Met v Met/Met	-47.363*	.019	-85.659	-8.985

			95% Confidence Interval	
RT Cost	Mean Difference	*p*	Lower	Upper
Val/Val v Val/Met	3.258	.762	-17.248	25.348
Val/Val v Met/Met	-11.024	.415	-36.908	15.698
Val/Met v Met/Met	-14.282	.187	-36.054	5.966

Congruent Trial ER	Mean Difference	*p*	Lower	Upper
Val/Val v Val/Met	0.010	.142	0.007	0.100
Val/Val v Met/Met	0.006	.350	-0.010	0.088
Val/Met v Met/Met	-0.003	.434	-0.038	0.008

			95% Confidence Interval	
Incongruent Trial ER	Mean Difference	*p*	Lower	Upper
Val/Val v Val/Met	0.050*	.041	0.007	0.100
Val/Val v Met/Met	0.036	.144	-0.010	0.088
Val/Met v Met/Met	-0.014	.244	-0.038	0.008

			95% Confidence Interval	
ER Cost	Mean Difference	*p*	Lower	Upper
Val/Val v Val/Met	0.040	.071	0.001	0.086
Val/Val v Met/Met	0.029	.182	-0.011	0.075
Val/Met v Met/Met	-0.011	.298	-0.031	0.008

*RT = response time, ER = error rate; * $p \leq .05$; + $p \leq .10$*

Table A.5 Summary of boostrapped pairwise comparisons for the effect of COMT Val158Met on response time and error rate for the dual task test

			95% Confidence Interval	
Single Trial RT	Mean Difference	*p*	Lower	Upper
Val/Val v Val/Met	-100.570*	.002	-163.157	-37.132
Val/Val v Met/Met	-84.963*	.026	-157.655	-8.898
Val/Met v Met/Met	15.607	.617	-45.119	72.743

			95% Confidence Interval	
Dual Trial RT	Mean Difference	*p*	Lower	Upper
Val/Val v Val/Met	-120.527*	.004	-200.525	-40.873
Val/Val v Met/Met	-141.768*	.002	-231.982	-50.877
Val/Met v Met/Met	-21.240	.533	-88.669	45.679

			95% Confidence Interval	
RT Cost	Mean Difference	*p*	Lower	Upper
Val/Val v Val/Met	-19.957	.394	-66.785	26.495
Val/Val v Met/Met	-56.804*	.020	-105.640	-9.491
Val/Met v Met/Met	-36.847+	.096	-79.919	6.239

			95% Confidence Interval	
Single Trial ER	Mean Difference	*p*	Lower	Upper
Val/Val v Val/Met	-0.034*	.027	-0.062	-0.004
Val/Val v Met/Met	-0.010	.565	-0.042	0.023
Val/Met v Met/Met	0.025+	.069	-0.003	0.050

			95% Confidence Interval	
Dual Trial ER	Mean Difference	*p*	Lower	Upper
Val/Val v Val/Met	-0.082*	.006	-0.140	-0.024
Val/Val v Met/Met	-0.043	.185	-0.106	0.021
Val/Met v Met/Met	0.039	.107	-0.009	0.087

			95% Confidence Interval	
ER Cost	Mean Difference	*p*	Lower	Upper
Val/Val v Val/Met	-0.048*	.009	-0.080	-0.015
Val/Val v Met/Met	-0.033	.150	-0.080	0.011
Val/Met v Met/Met	0.014	.483	-0.026	0.052

*RT = response time, ER = error rate; * $p \leq .05$; + $p \leq .10$*

Figures for DBH 444 G/A genotypic analysis

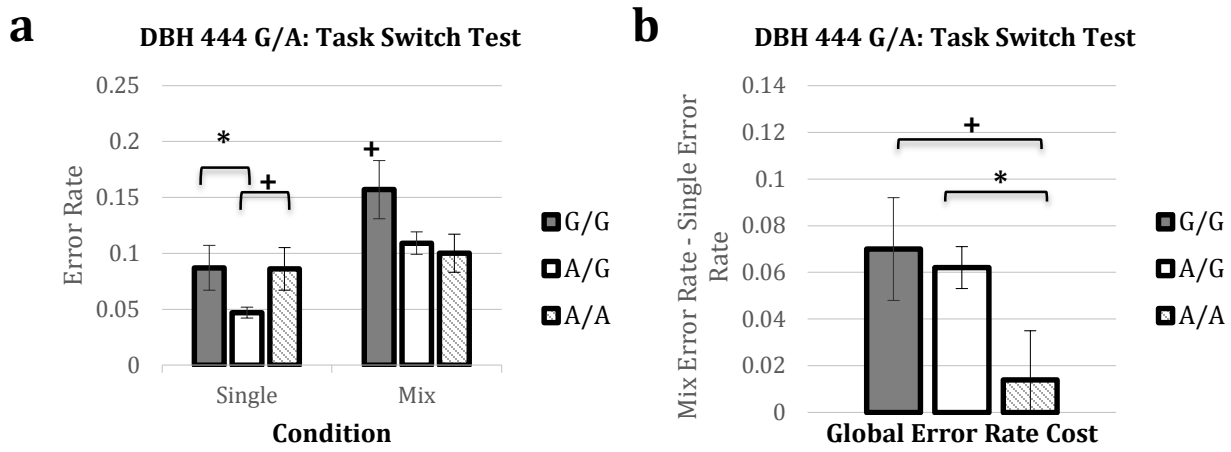


Figure A.1 Effect of the DBH 444 G/A SNP on error rate during the two conditions (i.e. single and mix) of the task switch test. **a)** There was a significant effect of genotype on error rate (ER) for the single condition, $F(2, 151) = 4.28$, $p = .016$. Bootstrapped pairwise comparisons: G/A vs G/G, mean difference = -.040, $p = .058$, 95% CI [-.084, -.002]; G/A vs A/A, mean difference = -.039, $p = .060$, 95% CI [-.080, -.002]. There was also marginal genotypic effect on ER in the mix condition, $F(2, 151) = 2.79$, $p = .064$. Bootstrapped pairwise comparisons: G/G vs G/A, mean difference = .048, $p = .083$, 95% CI [.003, .092]; G/G/ vs A/A, mean difference = .057, $p = .069$, 95% CI [.005, .109] **b)** There was a significant difference between the groups on a global error cost score (i.e. mix condition ER – single condition ER), $F(2, 151) = 3.34$, $p = .038$. Bootstrapped pairwise comparisons: A/A vs G/A, mean difference = -.048, $p = .031$, 95% CI [-.094, -.004]; A/A/ vs G/G group, mean difference = -.056, $p = .065$, 95% CI [-0.117, 0.00]. * $p \leq .05$; + $p \leq .10$

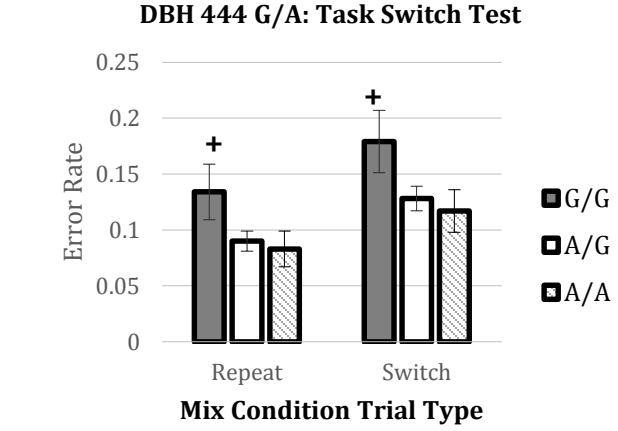


Figure A.2 Effect of the DBH 444 G/A SNP on error rate during the two trial types (i.e. repeat and switch) of the mix condition of the task switch test. There was a marginal genotypic effect on ER for repeat trials, $F(2, 151) = 2.60$, $p = .077$. Bootstrapped pairwise comparisons: G/G vs G/A, mean difference = .044, $p = .100$, 95% CI [-.005, .098]; G/G vs A/A, mean difference = .051, $p = .089$, 95% CI [-.007, .111]. There was also a marginal effect on ER for switch trials, $F(2, 151) = 2.61$, $p = .077$; G/G vs G/A, mean difference = .051, $p = .094$, 95% CI [-.006, .114]; G/G vs A/A, mean difference = .063, $p = .066$, 95% CI [-.002, .132]. + $p \leq .10$

Figures for DBH -1021 C/T allelic analysis

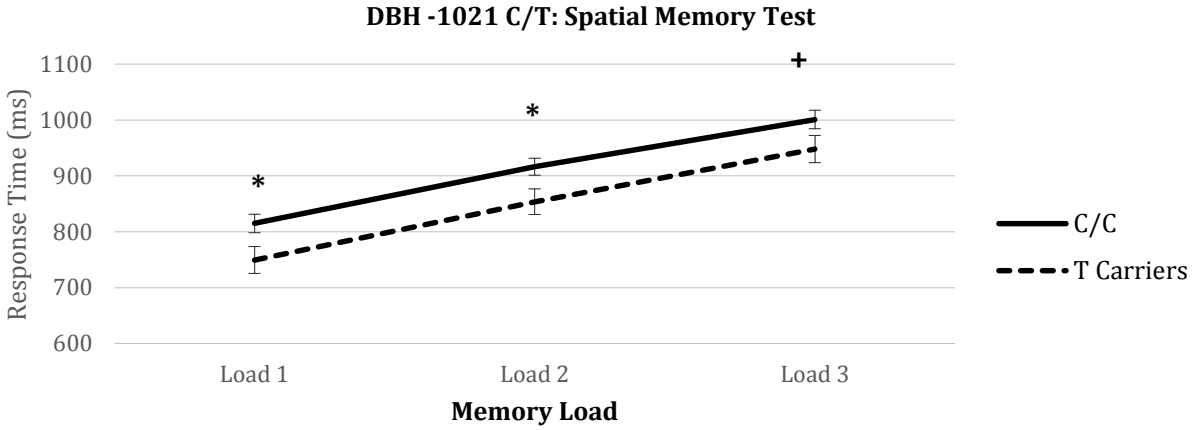


Figure A.3 Effect of the DBH -1021 C/T SNP on response time (RT) during the three memory loads of the spatial memory test (i.e. load 1, 2, and 3). T Carriers showed faster response times (RT) across all memory loads (i.e. 1, 2, and 3) on the spatial memory test: memory load 1 RT, $F(1, 170) = 5.14$, $p = .025$, mean difference = -65.43, $p = .029$, 95% CI [-122.79, -6.16]; memory load 2 RT, $F(1, 170) = 4.94$, $p = .028$, mean difference = -62.50, $p = .027$, 95% CI [-116.79, -7.09]; memory load 3 RT, $F(1, 170) = 2.34$, $p = .088$, mean difference = -52.98, $p = .083$, 95% CI [-112.40, 6.54]. * $p \leq .05$, + $p \leq .10$

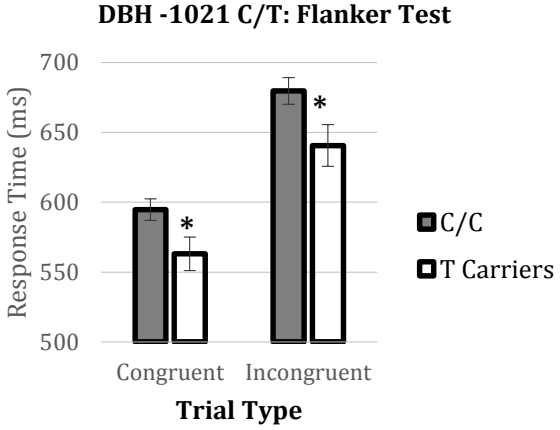


Figure A.4 Effect of the DBH -1021 C/T SNP on response time (RT) for the two trial types (i.e. congruent and incongruent) of the Flanker test. T Carriers showed faster response times (RT) on both trial types: Congruent, $F(1, 181) = 4.13$, $p = .044$, mean difference = -29.83, $p = .051$, 95% CI [-59.33, 0.50]; Incongruent, $F(1, 181) = 4.09$, $p = .045$, mean difference = -36.58, $p = .049$, 95% CI [-72.23, -0.61]. * $p \leq .05$

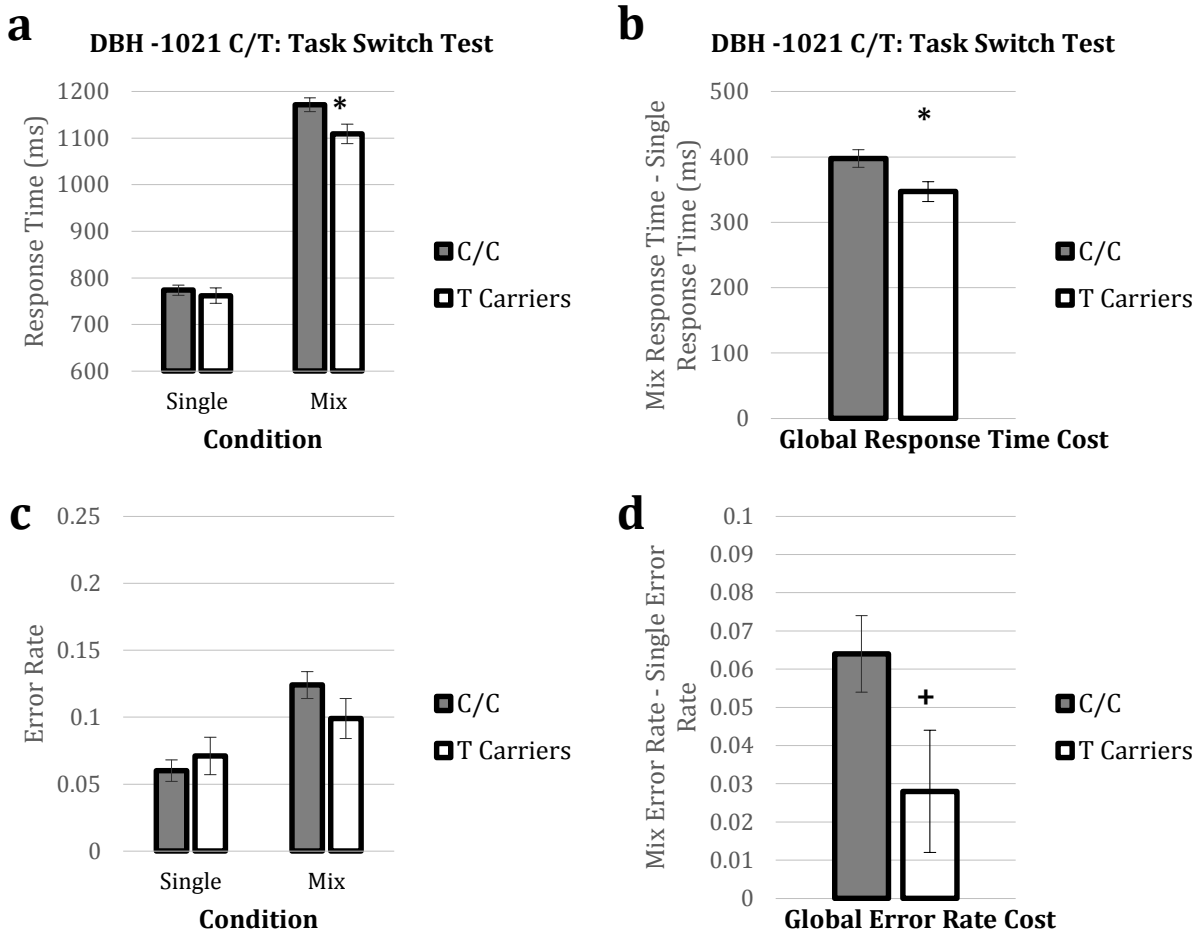


Figure A.5 Effect of the DBH -1021 C/T SNP on response time (RT) during the two conditions (i.e. single and mix) of the task switch test. **a)** No significant differences between the groups on response time (RT) for the single condition, $F(1, 151) < 1$, but the T Carriers were significantly faster during the mix condition $F(1, 151) = 6.20$, $p = .014$, mean difference = -62.63, $p = .016$, 95% CI [-113.36, -13.37] **b)** T Carriers had a significantly lower global RT cost, $F(1, 151) = 5.21$, $p = .024$, mean difference = -50.71, $p = .014$, 95% CI [-92.19, -9.81] **c)** No group differences on error rate (ER) for either the single condition, $F(1, 151) < 1$, or the mix condition, $F(1, 151) = 1.85$, $p = .176$ **d)** There was a marginal group difference on a global error rate cost score, $F(1, 151) = 3.88$, $p = .051$, with T Carriers showing a lower cost to the mix compared to the single condition in terms of error rate compared to the C/C group, mean difference = -.037, $p = .058$, 95% CI [-.075, .001]. * $p \leq .05$, + $p \leq .10$

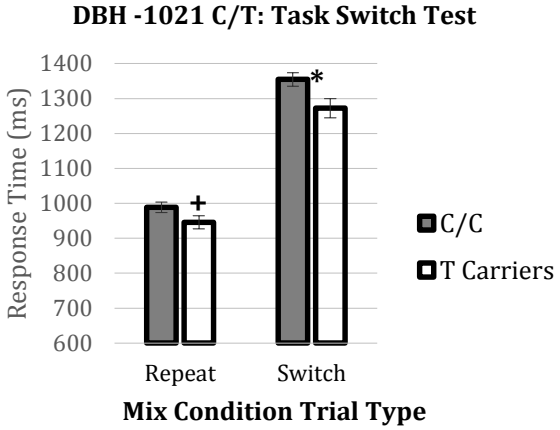


Figure A.6 Effect of the DBH -1021 C/T SNP on response time (RT) for the two trial types (i.e. repeat and switch) of the mix condition of the task switch test. **a)** T Carriers were marginally faster than C/C Homozygotes on RT for repeat trials, $F(1, 151) = 2.97$, $p = .087$, mean difference = -42.95, $p = .079$, 95% CI [-89.77, 6.16], and significantly faster on switch trials, $F(1, 151) = 6.28$, $p = .013$, mean difference = -82.31, $p = .017$, 95% CI [-146.96, -15.73]. * $p \leq .05$, + $p \leq .10$

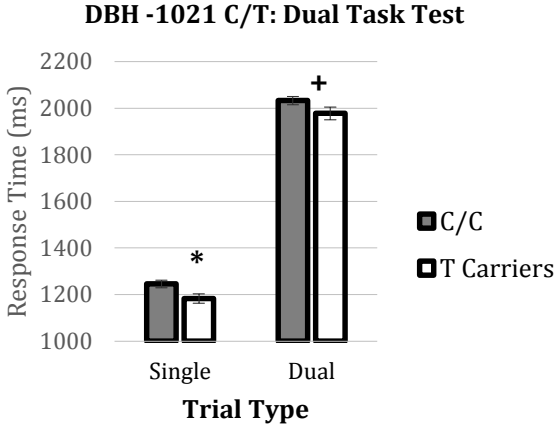


Figure A.7 Effect of the DBH -1021 C/T SNP on response time (RT) for the two trial types (i.e. single and dual) of dual task test. T Carriers showed significantly faster response times (RT) on both single trials, $F(1, 152) = 4.82$, $p = .030$, mean difference = -62.06, $p = .014$, 95% CI [-113.51, -13.48], and marginally on dual trials, $F(1, 152) = 2.77$, $p = .098$, mean difference = -55.11, $p = .091$, 95% CI [-118.77, 9.57]. * $p \leq .05$, + $p \leq .10$

Figures for DBH Combined allelic analysis

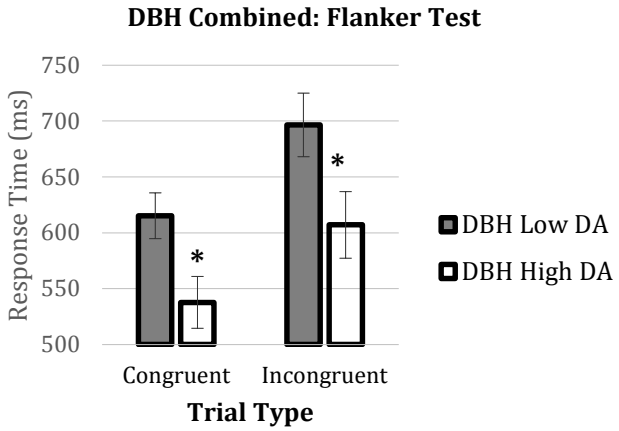


Figure A.8 Effect of the Combined DBH SNPs on response time (RT) for the two trial types (i.e. congruent and incongruent) of Flanker test. The DBH High DA group was significantly faster than the DBH Low DA group on both trial types: congruent, $F(1, 41) = 4.96$, $p = .031$, mean difference = -77.53, $p = .058$, 95% CI [-158.23, -3.81]; incongruent, $F(1, 41) = 5.11$, $p = .029$, mean difference = -89.48, $p = .045$, 95% CI [-179.08, -13.43]. * $p \leq .05$

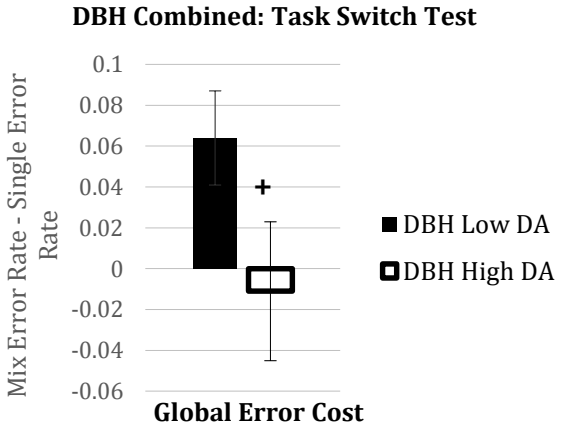


Figure A.9 Effect of the Combined DBH SNPs on error rate (ER) for the two conditions (i.e. single and mix) of task switch test. the DBH High DA group showed a marginally lower global ER cost (i.e. mix RT – Single RT), $F(1, 36) = 3.17$, $p = .083$, mean difference = -.074, $p = .094$, 95% CI [-.161, .005]. + $p \leq .10$

Figures for COMT Val158Met genotypic analysis

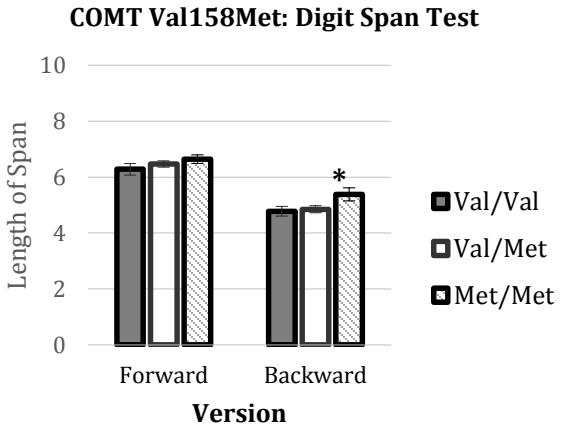


Figure A.10 Effect of the COMT Val158Met SNP on the length of span for the digit span test. forward span: $F(2, 189) = 1.05$, $p = .352$. backward span: $F(2, 189) = 2.45$, $p = .089$. Val/Val vs Val/Met, mean difference = 0.53, $p = .048$, 95% CI [0.02, 1.01]; Val/Val vs Met/Met, mean difference = 0.60, $p = .040$, 95% CI [0.04, 1.19]; Val/Met vs Met/Met, mean difference = .067, $p = .787$, 95% CI [-0.32, 0.47]. * $p \leq .05$

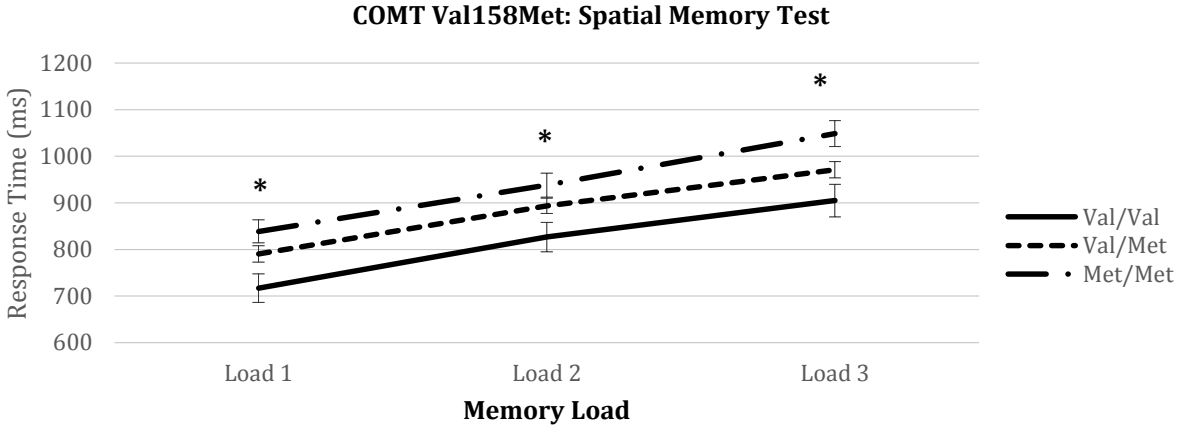


Figure A.11 Effect of COMT Val158Met on response time (RT) during the three memory loads of the spatial memory test: memory load 1, $F(2, 169) = 4.81$, $p = .009$; memory load 2, $F(2, 169) = 4.13$, $p = .018$; memory load 3, $F(2, 169) = 6.41$, $p = .002$. * $p \leq .05$

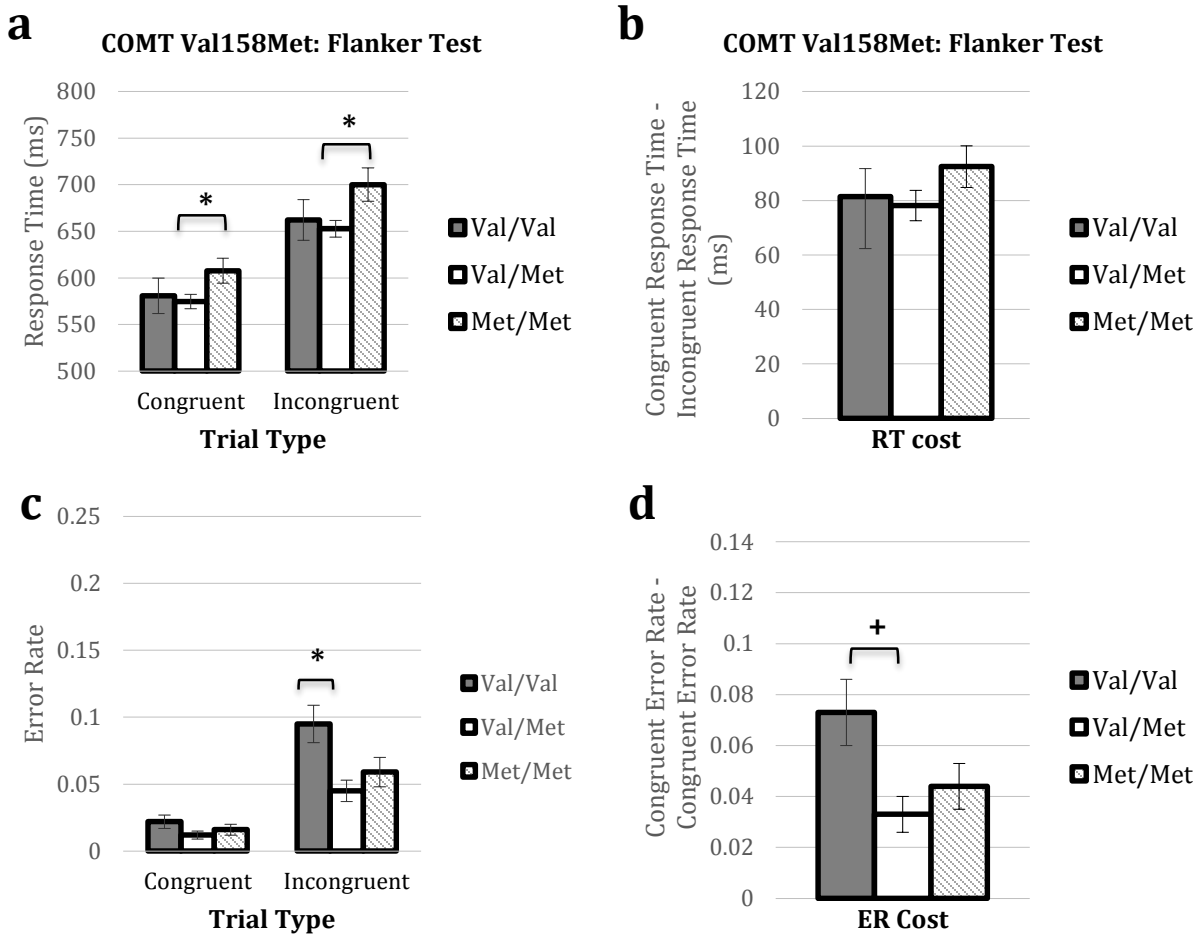


Figure A.12 Effect of COMT Val158Met on response time (RT) and error rate (ER) for the two trial types (i.e. congruent and incongruent) of Flanker test. **a)** Marginal effect on RT for congruent trials, $F(2, 180) = 2.55$, $p = .081$; significant effect on RT for incongruent trials, $F(2, 180) = 3.47$, $p = .033$ **b)** Non-significant effect on an RT cost score, $F(2, 180) = 1.15$, $p = .320$ **c)** Non-significant effect on ER for congruent trials, $F(2, 180) = 1.45$, $p = .238$; significant effect on ER for incongruent trials, $F(2, 180) = 4.63$, $p = .011$. **d)** Significant difference on ER cost, $F(2, 180) = 3.86$, $p = .023$. See Table A.4 for bootstrapped pairwise comparisons. * $p \leq .05$; + $p \leq .10$

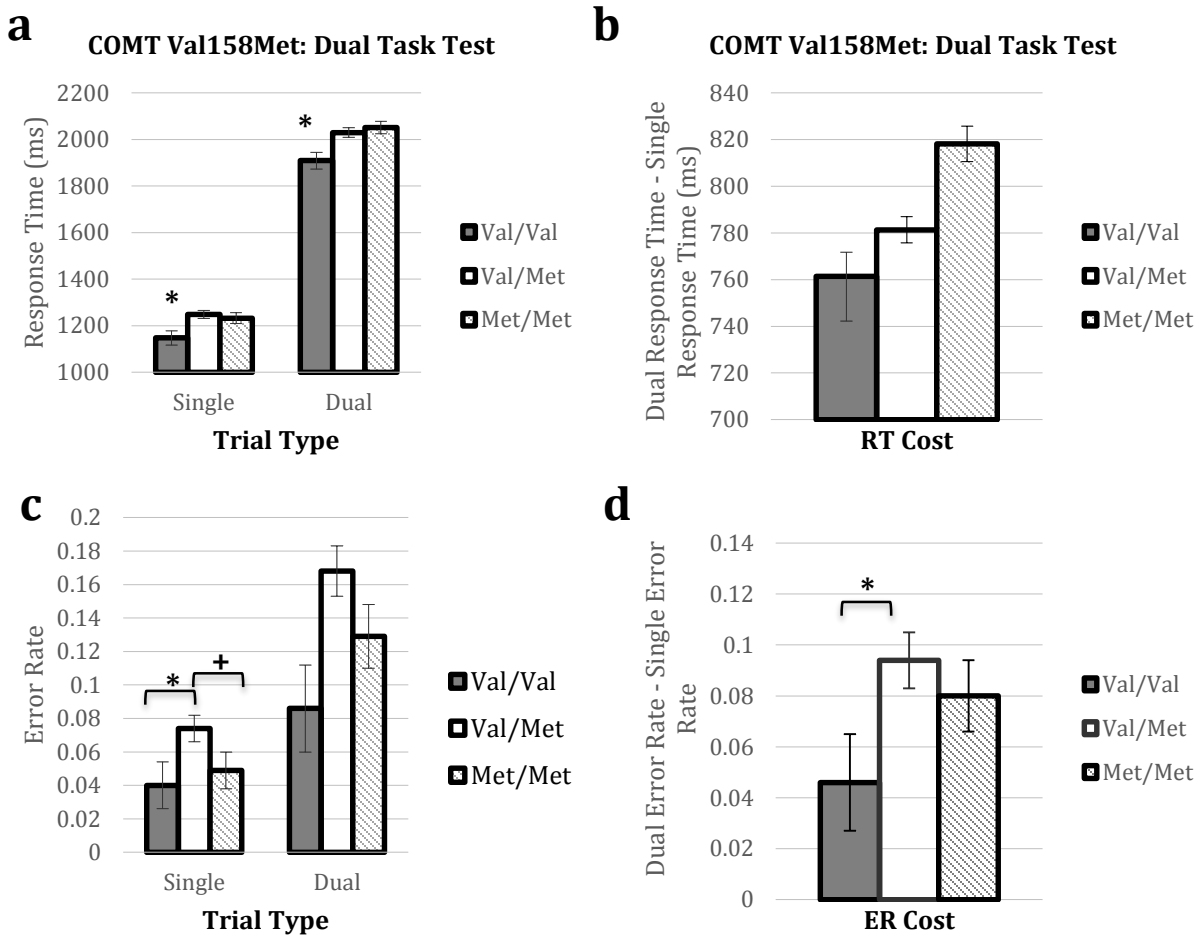


Figure A.13 Effect of the COMT Val158Met SNP on response time and error rate for the dual task test. **a)** There were significant differences between the genotypic groups on response time (RT) for both single, $F(2, 157) = 4.07$, $p = .019$, and dual trial types, $F(2, 151) = 5.50$, $p = .005$ **b) T**here were no no group differences on an RT cost score, $F(2, 151) = 2.16$, $p = .118$ **c)** There were differences between the groups on error rate (ER) for both single, $F(2, 157) = 2.99$, $p = .054$, and dual trial types, $F(2, 157) = 4.22$, $p = .017$ **d)** There was a marginal difference on an ER cost score, $F(2, 151) = 2.44$, $p = .091$. See Table A.5 for bootstrap pairwise comparisons. * $p \leq .05$; + $p \leq .10$

Figures for COMT C/G allelic analysis

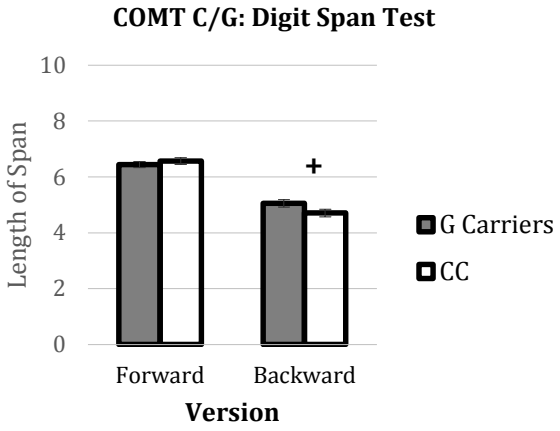


Figure A.14 Effect of COMT Val158Met SNP on length of span for both versions of the digit span test. There were no group differences on forward span, $F(1, 190) < 1$, but for backward span, G Carriers had marginally longer spans, $F(1, 190) = 3.36$, $p = .068$, mean difference = 0.34, $p = .068$, 95% CI [-0.03, 0.71].+ $p \leq .10$

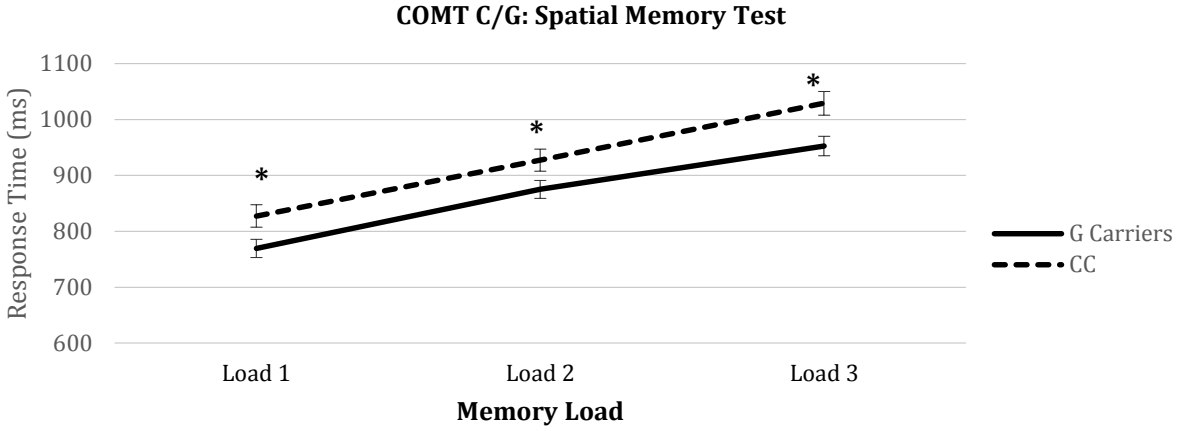


Figure A.15 Effect of the COMT C/G SNP on response time (RT) during the three memory loads of the spatial memory test (i.e. load 1, 2, and 3). G Carriers had significantly faster response times (RT) for all memory loads: memory load 1, $F(1, 170) = 5.00$, $p = .027$, mean difference = -57.87, $p = .029$, 95% CI [-109.13, -6.10]; memory load 2, $F(1, 170) = 4.21$, $p = .042$, mean difference = -52.28, $p = .045$, 95% CI [-103.64, -2.05]; memory load 3, $F(1, 170) = 7.71$, $p = .006$, mean difference = -76.46, $p = .010$, 95% CI [-134.05, -20.85].
* $p \leq .05$

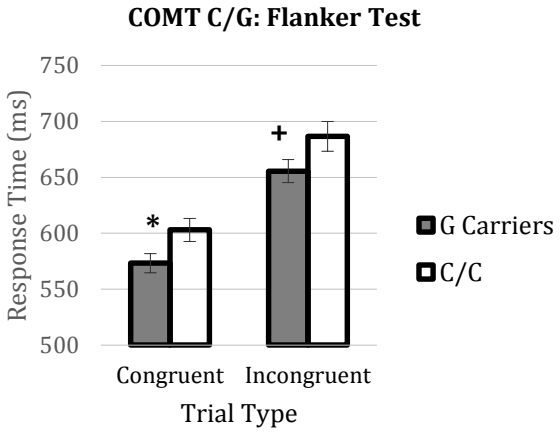


Figure A.16 Effect of the COMT C/G SNP on response time (RT) during the two trial types of the Flanker Test. G Carriers were significantly faster than C/C Homozygotes on congruent trials, $F(1, 181) = 5.22$, $p = .023$, mean difference = -29.78, $p = .024$, 95% CI [-56.53, -3.90], and marginally faster on incongruent trials, $F(1, 181) = 3.73$, $p = .055$, mean difference = -31.14, $p = .063$, 95% CI [-64.03, 1.35]. * $p \leq .05$; + $p \leq .10$

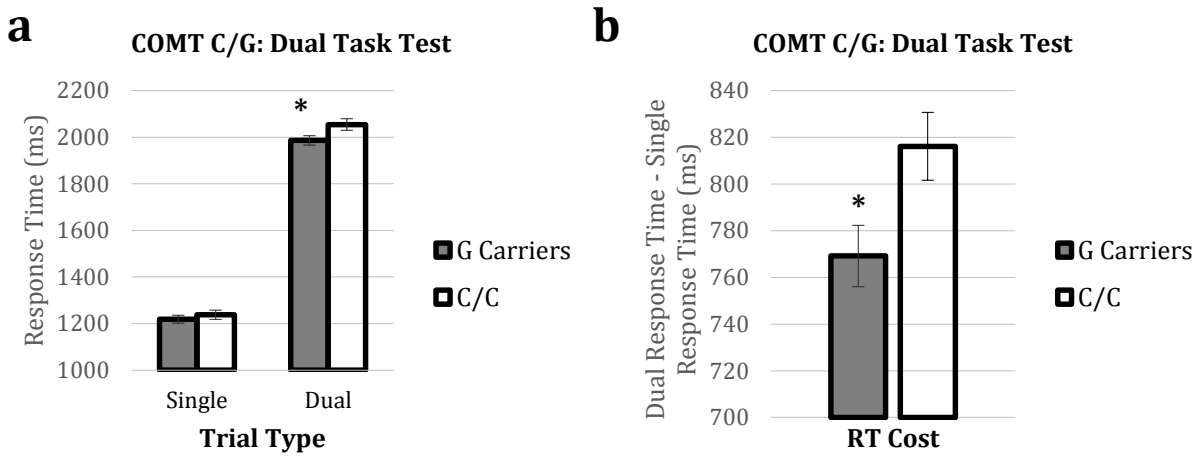


Figure A.17 Effect of the COMT C/G SNP on response time (RT) for the two trial types (i.e. single and dual) of dual task test. **a)** G Carriers were significantly faster on dual trials, $F(1, 152) = 4.86$, $p = .029$, mean difference = -66.91, $p = .029$, 95% CI [-126.78, -8.42], but there was no difference on single trials, $F(1, 152) < 1$ **b)** G Carriers also showed a significantly lower RT cost, $F(1, 152) = 5.71$, $p = .018$, mean difference = -46.95, $p = .018$, 95% CI [-84.77, -9.17]. * $p \leq .05$

Figures for COMT Combined analysis

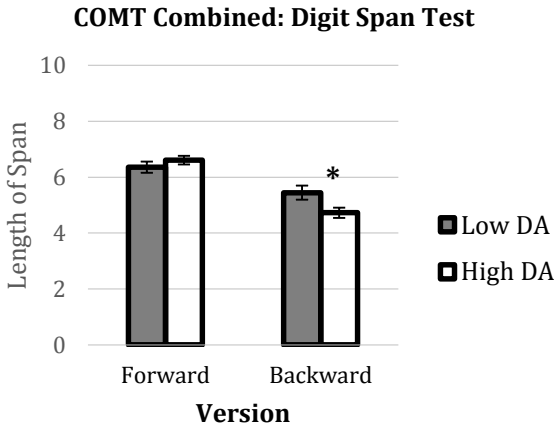


Figure A.18 Effect of the COMT Combined on the length of span for the digit span test. Forward span: $F(1, 76) < 1$. Backward span: $F(1, 76) = 5.63$, $p = .020$, mean difference = 0.72, $p = .018$, 95% CI [0.15, 1.31]. * $p \leq .05$

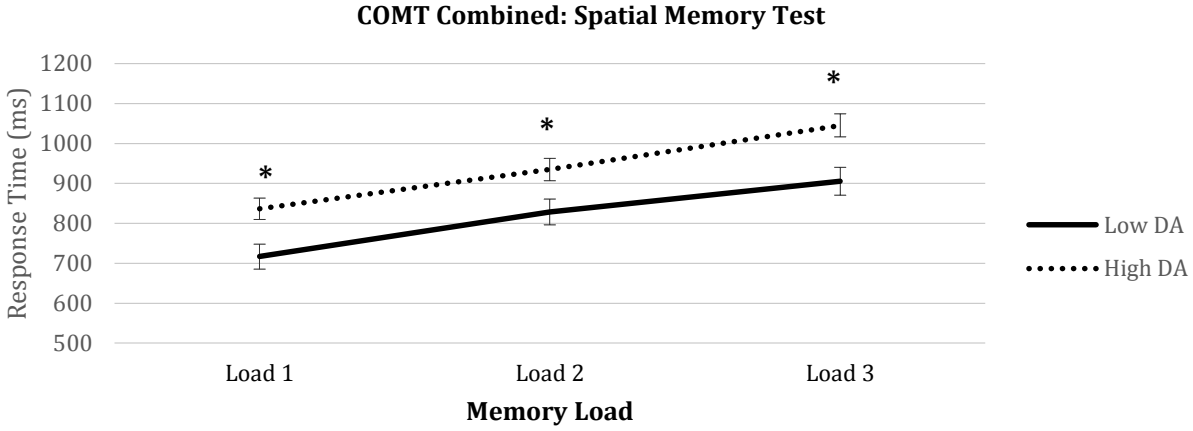


Figure A.19 Effect of COMT Combined on response time (RT) during the three memory loads of the spatial memory test (i.e. load 1, 2, and 3). The Low DA group had significantly faster response times (RT) for all memory loads: memory load 1, $F(1, 71) = 8.53$, $p = .005$, mean difference = -119.64, $p = .005$, 95% CI [-198.38, -43.13]; memory load 2, $F(1, 71) = 6.17$, $p = .015$, mean difference = -106.56, $p = .013$, 95% CI [-191.22, -26.50]; memory load 3, $F(1, 71) = 8.99$, $p = .004$, mean difference = -139.73, $p = .002$, 95% CI [-223.21, -54.82]. * $p \leq .05$

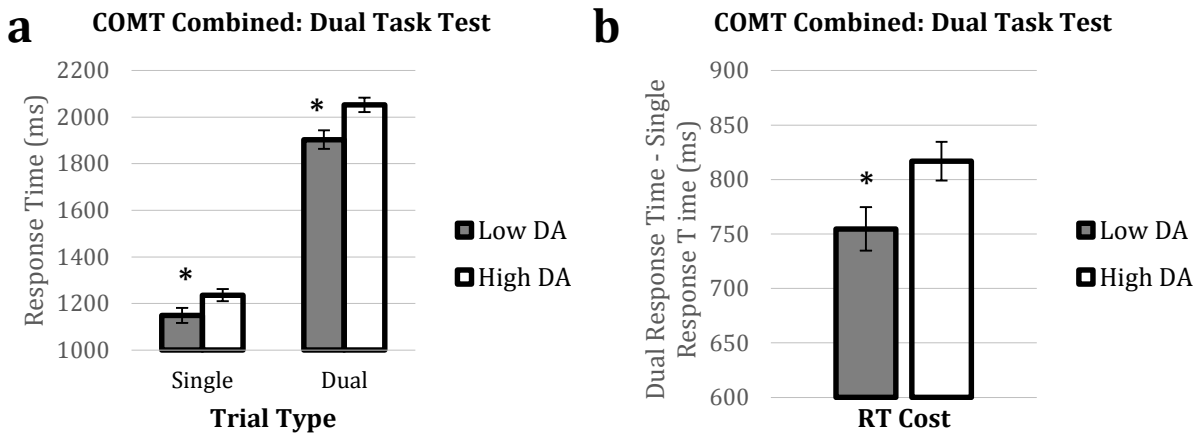


Figure A.20 Effect of COMT Combined on response time (RT) during the two trial types (i.e. single and dual) of the dual task test. **a)** The Low DA group was significantly faster for both single, $F(1, 68) = 3.99$, $p = .050$, mean difference = -87.08, $p = .040$, 95% CI [-166.76, -3.55], and dual trials, $F(1, 68) = 8.93$, $p = .004$, mean difference = -149.37, $p = .003$, 95% CI [-242.37, -55.75]. **b)** The Low DA group had a significantly lower RT, $F(1, 68) = 5.90$, $p = .018$, mean difference = -62.30, $p = .015$, 95% CI [-111.92, -15.29]. * $p \leq .05$

Appendix B: Growth Factor Genes Supplemental Figures and Tables

Tables for IGF1 G/A genotypic analysis

Table B.1 Summary of boostrapped pairwise comparisons for the effect of IGF1 G/A on response time for the spatial memory test

			95% Confidence Interval	
Memory Load 1	Mean Difference	*p*	Lower	Upper
A/A v A/G	59.762*	.033	5.322	113.116
A/A v G/G	-42.722	.399	-141.002	61.436
A/G v G/G	-102.484*	.044	-202.212	2.859

			95% Confidence Interval	
Memory Load 2	Mean Difference	*p*	Lower	Upper
A/A v A/G	35.726	.198	-18.349	89.657
A/A v G/G	-29.676	.587	-136.354	81.907
A/G v G/G	-65.403	.236	-171.661	46.394

			95% Confidence Interval	
Memory Load 3	Mean Difference	*p*	Lower	Upper
A/A v A/G	70.484*	.020	11.418	128.296
A/A v G/G	-50.734	.367	-162.373	64.806
A/G v G/G	-121.218*	.041	-235.265	-3.635

Table B.2 Summary of boostrapped pairwise comparisons for the effect of IGF1 G/A on error rate for the task switch test

			95% Confidence Interval	
Single Error Rate	Mean Difference	*p*	Lower	Upper
A/A v A/G	-0.010	.439	-0.037	0.016
A/A v G/G	-0.051	.147	-0.122	0.013
A/G v G/G	-0.041	.245	-0.114	0.023

Table B.3 Summary of ANCOVA analysis of the effect of BDNF Val66Met on the spatial memory test

	F(1, 171)	*p*
Memory Load 1		
Response Time	0.405	.525
Error Rate	1.258	.264
Memory Load 2		
Response Time	1.285	.259
Error Rate	2.404	.123
Memory Load 3		
Response Time	1.304	.255
Error Rate	0.298	.546

Table B.4 Summary of ANCOVA analysis of the effect of the growth factor genes combined on the spatial memory test

	F(1, 18)	*p*
Memory Load 1		
Response Time	0.473	0.500
Error Rate	1.100	0.308
Memory Load 2		
Response Time	0.042	0.840
Error Rate	0.981	0.335
Memory Load 3		
Response Time	0.630	0.438
Error Rate	0.016	0.899

Figures for IGF1 G/A genotypic analysis

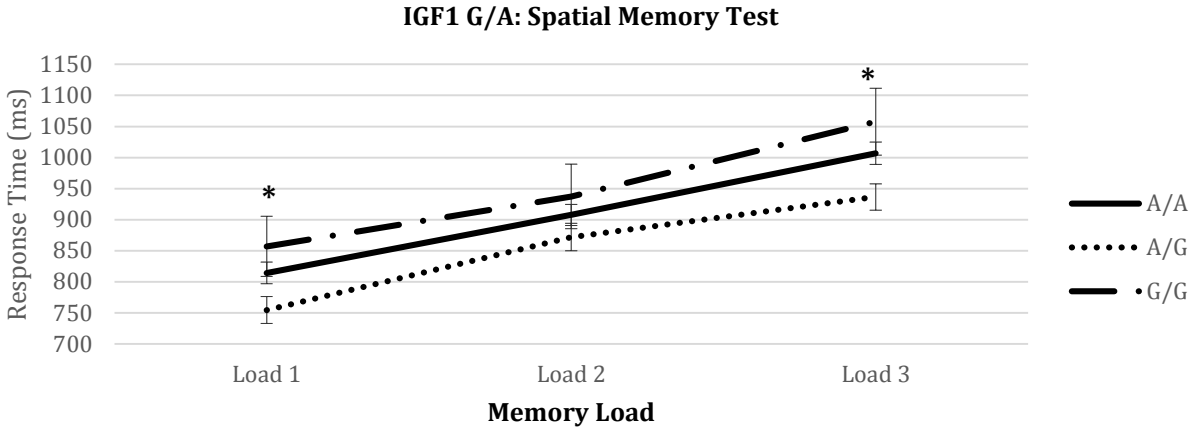


Figure B.1 Effect of the IGF1 G/A SNP on response time (RT) during the three memory loads of the spatial memory test (i.e. load 1, 2, and 3). There was a significant difference between the groups on response time for memory load 1, $F(2, 169) = 3.46$, $p = .034$, and memory load 3, $F(2, 169) = 4.30$, $p = .015$. Although the differences between genotypic groups were not significant for memory load 2, $F(2, 169) = 1.35$, $p = .262$, the trend was the same. Bootstrapped pairwise comparisons are reported in Table B.1. * $p \leq .05$

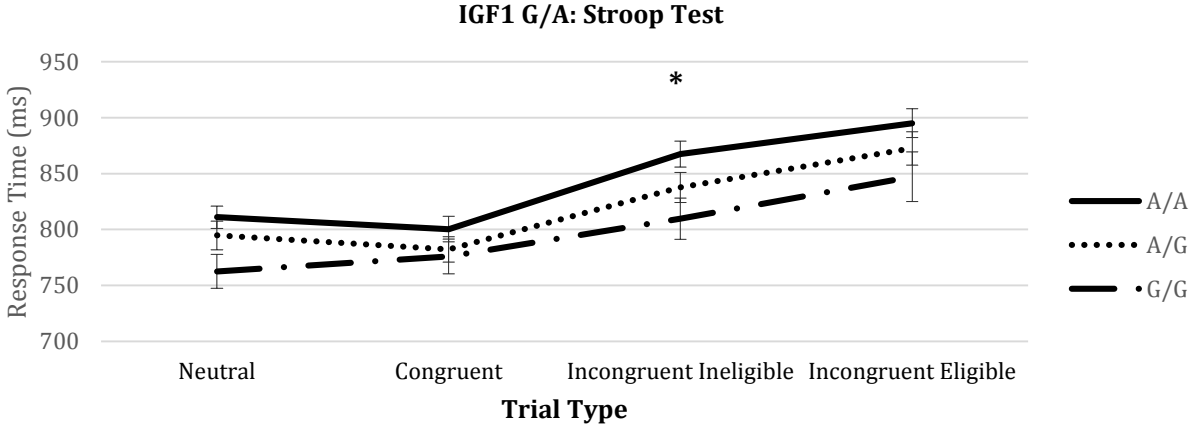


Figure B.2 Effect of the IGF1 G/A SNP on response time (RT) during the four trial types of the Stroop test. There was a significant group difference on RT for the incongruent-ineligible trial types, $F(2, 151) = 3.08$, $p = .049$, with the A/A group showing the slowest RTs. Bootstrapped pairwise comparisons: A/A vs G/A, mean difference = 29.74, $p = .091$, 95% CI [-5.21, 63.73], A/A vs G/G, mean difference = 57.71, $p = .007$, 95% CI [14.95, 100.35], G/A vs G/G, mean difference = 27.97, $p = .213$, 95% CI [-17.31, 72.64]. * $p \leq .05$

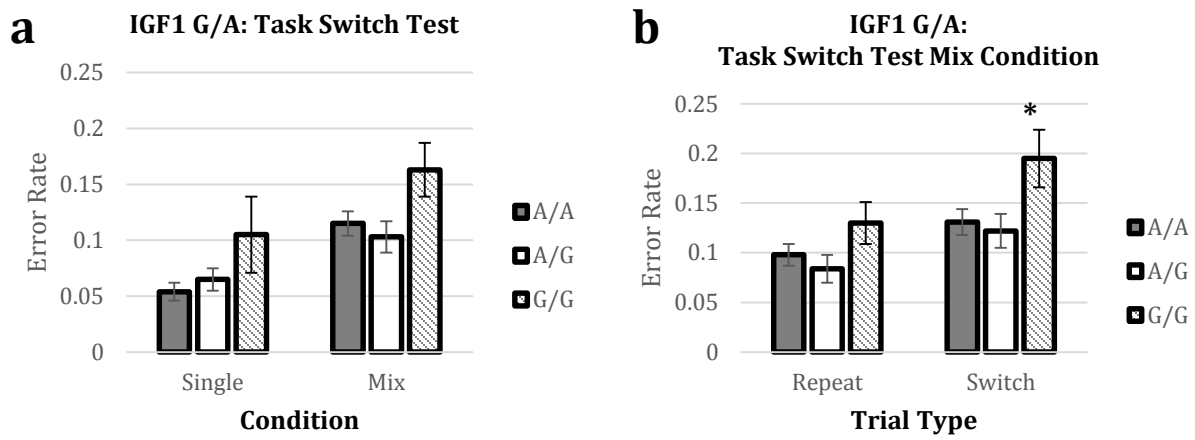


Figure B.3 Effect of the IGF1 G/A SNP on error rate (ER) for the task switch test. **a)** There was a marginal difference on error rate (ER) in the single condition, $F(2, 151) = 2.72$, $p = .069$. Bootstrapped pairwise comparisons: A/A vs G/A, mean difference = -0.01, $p = .439$, 95% CI [-0.04, 0.02]; A/A vs G/G, mean difference = -0.05, $p = .147$, 95% CI [-0.12, 0.01]; G/A vs G/G, mean difference = -0.04, $p = .245$, 95% CI [-0.11, 0.02]. There were no differences in ER in the mix Condition, $F(2, 151) = 2.14$, $p = .121$. **b)** There were no differences on error rate (ER) for repeat trials, $F(2, 151) = 1.44$, $p = .241$, but there was a marginal difference on ER for switch trials, $F(2, 151) = 2.64$, $p = .074$. Bootstrapped pairwise comparisons: A/A vs G/A, mean difference = .008, $p = .695$, 95% CI [-.033, .048]; A/A vs G/G group, mean difference = .064, $p = .046$, 95% CI [-.130, -.002]; G/A vs G/G group, mean difference = -.073, $p = .032$, 95% CI [-.140, -.007] * $p \leq .05$

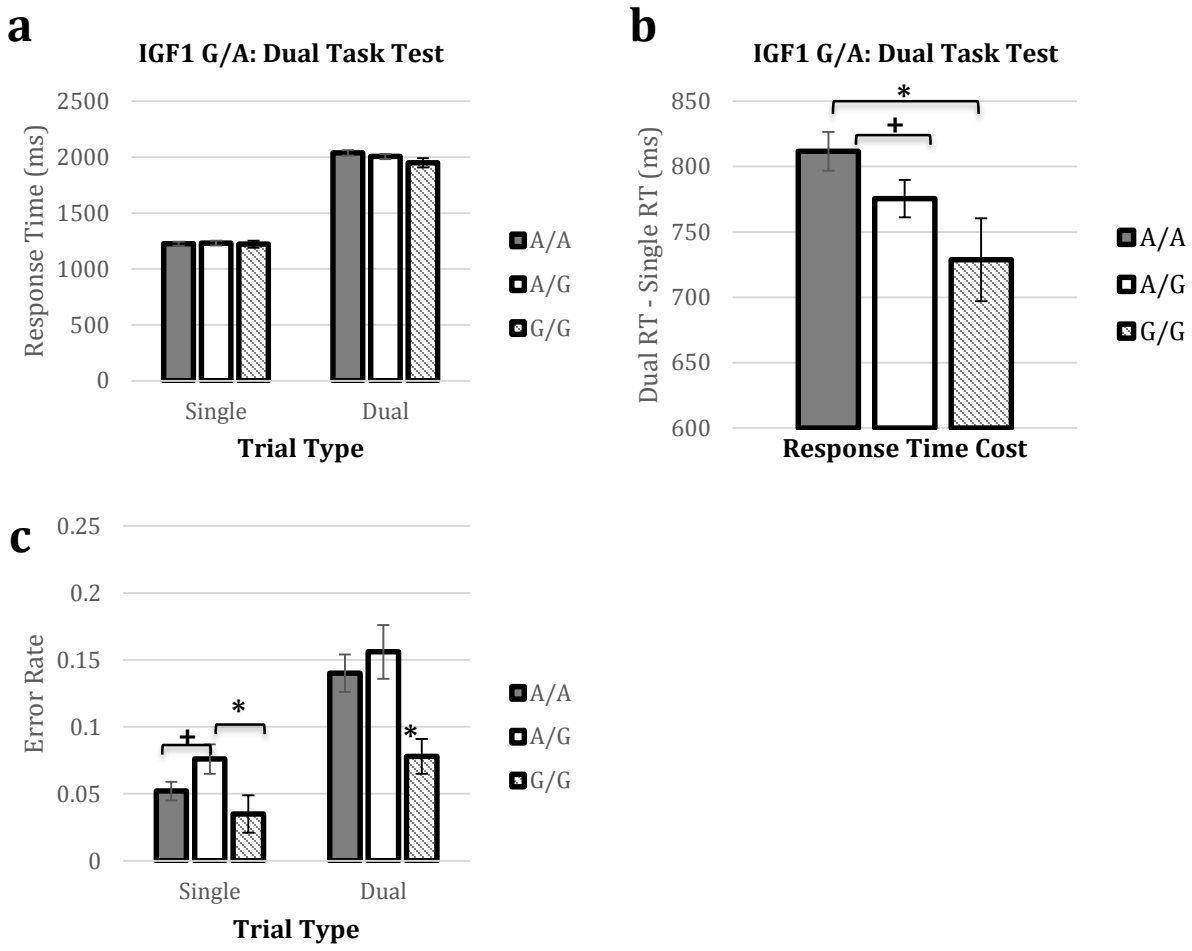


Figure B.4 Effect of the IGF1 G/A SNP on the dual task test. **a)** There were no significant differences between the genotypic groups on response time (RT) for either the single, $F(2, 150) < 1$, or dual trial types, $F(2, 150) = 1.69$, $p = .189$ **b)** There was a significant difference between the groups on a RT cost score, $F(2, 150) = 3.87$, $p = .023$. Bootstrapped pairwise comparisons: A/A vs G/A, mean difference = 36.33, $p = .078$, 95% CI [-3.23, 16.12]; A/A vs G/G, mean difference = 82.95, $p = .016$, 95% CI [15.70, 152.60], G/A vs G/G, mean difference = 34.06, $p = .171$, 95% CI [-19.32, 113.72] **c)** There was also a significant difference between the genotypic groups on error rate for single trials, $F(2, 150) = 3.05$, $p = .050$. Bootstrapped pairwise comparisons: A/A vs G/A, mean difference = -.023, $p = .069$, 95% CI [-.048, .002]; A/A vs G/G, mean difference = .017, $p = .268$, 95% CI [-.016, .046]; G/A vs G/G, mean difference = .040, $p = .020$, 95% CI [.005, .072]. There was also a marginal difference for error rate on dual trials, $F(2, 150) = 2.42$, $p = .092$. Bootstrapped pairwise comparisons: A/A vs G/A, mean difference = -.016, $p = .509$, 95% CI [-.061, .031]; A/A vs G/G group, mean difference = .062, $p = .003$, 95% CI [.026, .100]; G/A vs G/G, mean difference = .078, $p = .004$, 95% CI [.029, .125]. * $p \leq .05$, + $p \leq .10$

Figures for BDNF Val66Met

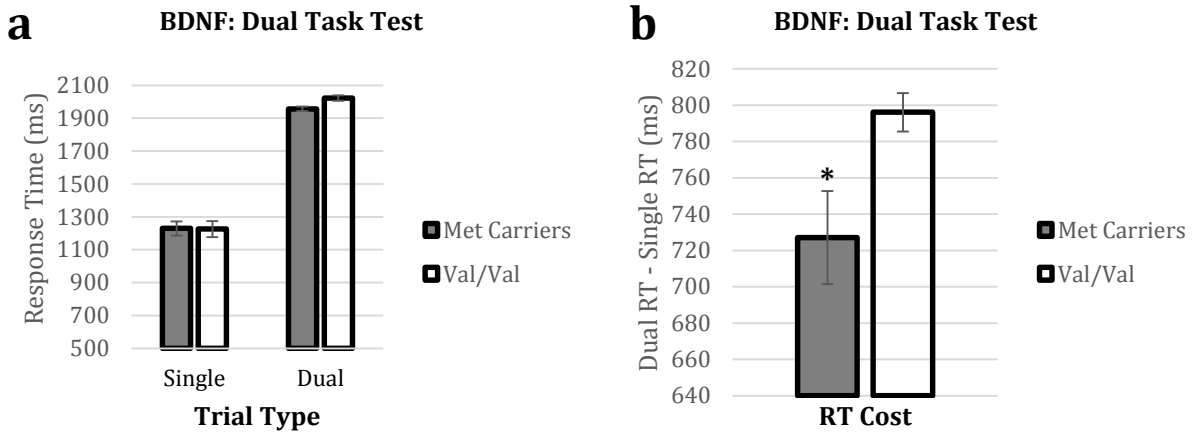


Figure B.5 Effect of the BDNF Val66Met on response time (RT) for the dual task test. **a)** There were no group differences RT for either single, $F(1, 153) < 1$, or dual trials, $F(1, 153) = 1.59$, $p = .209$ **b)** The groups did significantly differ on an RT cost score, $F(1, 153) = 4.34$, $p = .039$, with the Met Carriers showing a lower RT cost compared to the Val/Val Homozygotes, mean difference = -69.12, $p = .011$, 95% CI [-120.74, -13.84].
* $p \leq .05$

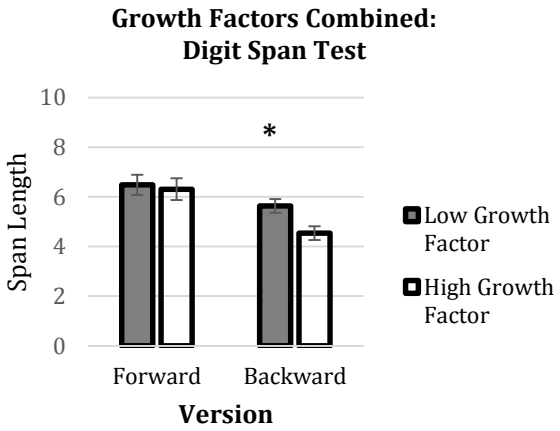


Figure B.6 Effect of the BDNF Val66Met on response time (RT) for the dual task test. There were no significant differences between the groups for forward span, $F(1, 25) < 1$, but the low GF group performed significantly better on the backward span, $F(1, 25) = 4.60$, $p = .042$, mean difference = 1.09, $p = .042$, 95% CI [-0.04, 1.99].

References

Aalto, S., Bruck, A., Laine, M., Nagren, K., & Rinne, J. (2005). Frontal and Temporal Dopamine Release during Working Memory and Attention Tasks in Healthy Humans: a Positron Emission Tomography Study Using the High-Affinity Dopamine D2 Receptor Ligand [11C]FLB 457. *Journal of Neuroscience*, *25*(10), 2471–2477. http://doi.org/10.1523/JNEUROSCI.2097-04.2005

Aalto, S., Bruck, A., Laine, M., Nagren, K., & Rinne, J. (2005). Frontal and Temporal Dopamine Release during Working Memory and Attention Tasks in Healthy Humans: a Positron Emission Tomography Study Using the High-Affinity Dopamine D2 Receptor Ligand [11C]FLB 457. *Journal of Neuroscience*, *25*(10), 2471–2477. http://doi.org/10.1523/JNEUROSCI.2097-04.2005

Aleman, A., & Torres-Alemán, I. (2009). Circulating insulin-like growth factor I and cognitive function: neuromodulation throughout the lifespan. *Progress in Neurobiology*, *89*(3), 256–265. http://doi.org/10.1016/j.pneurobio.2009.07.008

Aleman, A., Verhaar, H. J., Haan, E. H. D., Vries, W. R. D., Samson, M. M., Drent, M. L., … Koppeschaar, H. P. (1999). Insulin-like growth factor-I and cognitive function in healthy older men. *Journal of Clinical Endocrinology & Metabolism*, *84*(2), 471–5.

Al-Zahrani, A., Sandhu, M. S., Luben, R. N., Thompson, D., Baynes, C., Pooley, K. A., … Dunning, A. M. (2006). IGF1 and IGFBP3 tagging polymorphisms are associated with circulating levels of IGF1, IGFBP3 and risk of breast cancer. *Human Molecular Genetics*, *15*(1), 1–10. http://doi.org/10.1093/hmg/ddi398

Arwert, L. I., Deijen, J. B., & Drent, M. L. (2005). The relation between insulin-like growth factor I levels and cognition in healthy elderly: a meta-analysis. *Growth Hormone & IGF Research: Official Journal of the Growth Hormone Research Society and the International IGF Research Society*, *15*(6), 416–422. http://doi.org/10.1016/j.ghir.2005.09.001

Axelrod, J., & Tomchick, R. (1958). Enzymatic O-methylation of epinephrine and other catechols. *The Journal of Biological Chemistry*, *233*(3), 702–705.

Bäckman, L., & Farde, L. (2001). Dopamine and cognitive functioning: brain imaging findings in Huntington's disease and normal aging. *Scand J Psychol*, *42*(3), 287–96.

Bäckman, L., Ginovart, N., Dixon, R. A., Wahlin, T. B., Wahlin, A., Halldin, C., & Farde, L. (2000). Age-related cognitive deficits mediated by changes in the striatal dopamine system. *The American Journal of Psychiatry*, *157*(4), 635–637. http://doi.org/10.1176/ajp.157.4.635

Bäckman, L., Nyberg, L., Lindenberger, U., Li, S.-C., & Farde, L. (2006). The correlative triad among aging, dopamine, and cognition: current status and future prospects. *Neurosci Biobehav Rev*, *30*(6), 791–807. http://doi.org/10.1016/j.neubiorev.2006.06.005

Baddeley, A. (1992). Working memory. *Science*, *255*(5044), 556.

Baddeley, A. (1996). The fractionation of working memory. *Proc Natl Acad Sci USA*, *93*(24), 13468–72.

Baddeley, A. (2010). Working memory. *Curr Biol*, *20*(4), R136–40. http://doi.org/10.1016/j.cub.2009.12.014

Barnes, D. E., Yaffe, K., Satariano, W. A., & Tager, I. B. (2003). A longitudinal study of cardiorespiratory fitness and cognitive function in healthy older adults. *J Am Geriatr Soc*, *51*(4), 459–65.

Bellar, D., Glickman, E. L., Juvancic-Heltzel, J., & Gunstad, J. (2011). Serum insulin like growth factor-1 is associated with working memory, executive function and selective attention in a sample of healthy, fit older adults. *Neuroscience*, *178*, 133–137. http://doi.org/10.1016/j.neuroscience.2010.12.023

Bellgrove, M. A., & Mattingley, J. B. (2008). Molecular genetics of attention. *Ann N Y Acad Sci*, *1129*, 200–12. http://doi.org/10.1196/annals.1417.013

Berchtold, N. C., Castello, N., & Cotman, C. W. (2010). Exercise and time-dependent benefits to learning and memory. *Neuroscience*, *167*(3), 588–597. http://doi.org/10.1016/j.neuroscience.2010.02.050

Berchtold, N. C., Kesslak, J. P., Pike, C. J., Adlard, P. A., & Cotman, C. W. (2001). Estrogen and exercise interact to regulate brain-derived neurotrophic factor mRNA and protein expression in the hippocampus. *The European Journal of Neuroscience*, *14*(12), 1992–2002.

Bherer, L., Erickson, K. I., & Liu-Ambrose, T. (2013). A Review of the Effects of Physical Activity and Exercise on Cognitive and Brain Functions in Older Adults. *Journal of Aging Research*, *2013*, 1–8. http://doi.org/10.1155/2013/657508

Black, J. E., Isaacs, K. R., Anderson, B. J., Alcantara, A. A., & Greenough, W. T. (1990). Learning causes synaptogenesis, whereas motor activity causes angiogenesis, in cerebellar cortex of adult rats. *Proc Natl Acad Sci USA*, *87*(14), 5568–72.

Bondy, C. A., & Lee, W.-H. (1993). Patterns of Insulin-like Growth Factor and IGF Receptor Gene Expression in the Brain: Functional Implications. *Annals of the New York Academy of Sciences*, *692*(1 The Role of I), 33–43. http://doi.org/10.1111/j.1749-6632.1993.tb26203.x

Botvinick, M., Nystrom, L. E., Fissell, K., Carter, C. S., & Cohen, J. D. (1999). Conflict monitoring versus selection-for-action in anterior cingulate cortex. *Nature*, *402*(6758), 179–81. http://doi.org/10.1038/46035

Bouchard, T. J., Lykken, D. T., McGue, M., Segal, N. L., & Tellegen, A. (1990). Sources of human psychological differences: the Minnesota Study of Twins Reared Apart. *Science*, *250*(4978), 223–8.

Bourdélat-Parks, B. N., Anderson, G. M., Donaldson, Z. R., Weiss, J. M., Bonsall, R. W., Emery, M. S., … Weinshenker, D. (2005). Effects of dopamine beta-hydroxylase genotype and disulfiram inhibition on catecholamine homeostasis in mice. *Psychopharmacology*, *183*(1), 72–80. http://doi.org/10.1007/s00213-005-0139-8

Brink, J. M., & McDowd, J. M. (1999). Aging and selective attention: an issue of complexity or multiple mechanisms? *J Gerontol B Psychol Sci Soc Sci*, *54*(1), P30–3.

Brozoski, T. J., Brown, R. M., Rosvold, H. E., & Goldman, P. S. (1979). Cognitive deficit caused by regional depletion of dopamine in prefrontal cortex of rhesus monkey. *Science*, *205*(4409), 929–32.

Bueller, J. A., Aftab, M., Sen, S., Gomez-Hassan, D., Burmeister, M., & Zubieta, J.-K. (2006). BDNF Val66Met allele is associated with reduced hippocampal volume in healthy subjects. *Biological Psychiatry*, *59*(9), 812–815. http://doi.org/10.1016/j.biopsych.2005.09.022

Cabeza, R. (2001). Cognitive neuroscience of aging: contributions of functional neuroimaging. *Scand J Psychol*, *42*(3), 277–86.

Cabeza, R. (2001). Functional Neuroimaging of Cognitive Aging. In R. Cabeza & A. Kingstone (Eds.), *Handbook of functional neuroimaging of cognition* (1st ed., pp. 331–377). Cambridge, MA: MIT Press.

Cabeza, R. (2002). Hemispheric asymmetry reduction in older adults: the HAROLD model. *Psychology and Aging*, *17*(1), 85–100.

Carro, E., nez, A. N., Busiguina, S., & Torres-Aleman, I. (2000). Circulating insulin-like growth factor I mediates effects of exercise on the brain. *J Neurosci*, *20*(8), 2926–33.

Carro, E., Trejo, J. L., Busiguina, S., & Torres-Aleman, I. (2001). Circulating insulin-like growth factor I mediates the protective effects of physical exercise against brain insults of different etiology and anatomy. *J Neurosci*, *21*(15), 5678–84.

Cheng, C. M., Mervis, R. F., Niu, S.-L., Salem, N., Jr, Witters, L. A., Tseng, V., … Bondy, C. A. (2003). Insulin-like growth factor 1 is essential for normal dendritic growth. *Journal of Neuroscience Research*, *73*(1), 1–9. http://doi.org/10.1002/jnr.10634

Chen, J., Lipska, B. K., Halim, N., Ma, Q. D., Matsumoto, M., Melhem, S., … Weinberger, D. R. (2004). Functional analysis of genetic variation in catechol-O-methyltransferase (COMT): effects on mRNA, protein, and enzyme activity in postmortem human brain. *Am J Hum Genet*, *75*(5), 807–21. http://doi.org/10.1086/425589

Chen, T., & Li, D. (2007). The roles of working memory updating and processing speed in mediating age-related differences in fluid intelligence. *Aging, Neuropsychology, & Cognition*, *14*(6), 631–46. http://doi.org/10.1080/13825580600987660

Colcombe, S. J., Erickson, K. I., Raz, N., Webb, A. G., Cohen, N. J., Mcauley, E., & Kramer, A. F. (2003). Aerobic fitness reduces brain tissue loss in aging humans. *J Gerontol A Biol Sci Med Sci*, *58*(2), 176–80.

Colcombe, S. J., Erickson, K. I., Scalf, P. E., Kim, J. S., Prakash, R., Mcauley, E., … Kramer, A. F. (2006). Aerobic exercise training increases brain volume in aging humans. *J Gerontol A Biol Sci Med Sci*, *61*(11), 1166–70.

Colcombe, S. J., Kramer, A. F., Erickson, K. I., & Scalf, P. (2005). The implications of cortical recruitment and brain morphology for individual differences in inhibitory function in aging humans. *Psychology and Aging*, *20*(3), 363–75. http://doi.org/10.1037/0882-7974.20.3.363

Colcombe, S. J., Kramer, A. F., Erickson, K. I., Scalf, P., Mcauley, E., Cohen, N. J., … Elavsky, S. (2004). Cardiovascular fitness, cortical plasticity, and aging. *Proc Natl Acad Sci USA*, *101*(9), 3316–21. http://doi.org/10.1073/pnas.0400266101

Colcombe, S. J., Kramer, A. F., Mcauley, E., Erickson, K. I., & Scalf, P. (2004). Neurocognitive aging and cardiovascular fitness: recent findings and future directions. *J Mol Neurosci*, *24*(1), 9–14. http://doi.org/10.1385/JMN:24:1:009

Colcombe, S., & Kramer, A. F. (2003). Fitness effects on the cognitive function of older adults: a meta-analytic study. *Psychol Sci*, *14*(2), 125–30.
Cools, R., & Robbins, T. W. (2004). Chemistry of the adaptive mind. *Philosophical Transactions. Series A, Mathematical, Physical, and Engineering Sciences*, *362*(1825), 2871–2888. http://doi.org/10.1098/rsta.2004.1468
Cooper, J. (2003). *The biochemical basis of neuropharmacology* (8th ed.). Oxford ;;New York: Oxford University Press.
Cotman, C. W., & Berchtold, N. C. (2002). Exercise: a behavioral intervention to enhance brain health and plasticity. *Trends Neurosci*, *25*(6), 295–301.
Cotman, C. W., Berchtold, N. C., & Christie, L.-A. (2007). Exercise builds brain health: key roles of growth factor cascades and inflammation. *Trends Neurosci*, *30*(9), 464–72. http://doi.org/10.1016/j.tins.2007.06.011
Creer, D. J., Romberg, C., Saksida, L. M., van Praag, H., & Bussey, T. J. (2010). Running enhances spatial pattern separation in mice. *Proceedings of the National Academy of Sciences*, *107*(5), 2367–2372. http://doi.org/10.1073/pnas.0911725107
Cubells, J. F., Kammen, D. P. van, Kelley, M. E., Anderson, G. M., O'Connor, D. T., Price, L. H., … Gelernter, J. (1998). Dopamine beta-hydroxylase: two polymorphisms in linkage disequilibrium at the structural gene DBH associate with biochemical phenotypic variation. *Hum Genet*, *102*(5), 533–40.
Cubells, J. F., & Zabetian, C. P. (2004). Human genetics of plasma dopamine beta-hydroxylase activity: applications to research in psychiatry and neurology. *Psychopharmacology*, *174*(4), 463–76. http://doi.org/10.1007/s00213-004-1840-8
Deary, I. J., Wright, A. F., Harris, S. E., Whalley, L. J., & Starr, J. M. (2004). Searching for genetic influences on normal cognitive ageing. *Trends Cogn Sci (Regul Ed)*, *8*(4), 178–84. http://doi.org/10.1016/j.tics.2004.02.008
Deeny, S. P., Poeppel, D., Zimmerman, J. B., Roth, S. M., Brandauer, J., Witkowski, S., … Hatfield, B. D. (2008). Exercise, APOE, and working memory: MEG and behavioral evidence for benefit of exercise in epsilon4 carriers. *Biological Psychology*, *78*(2), 179–87. http://doi.org/10.1016/j.biopsycho.2008.02.007
Diatchenko, L., Slade, G. D., Nackley, A. G., Bhalang, K., Sigurdsson, A., Belfer, I., … Maixner, W. (2005). Genetic basis for individual variations in pain perception and the development of a chronic pain condition. *Human Molecular Genetics*, *14*(1), 135–43. http://doi.org/10.1093/hmg/ddi013
Dik, M. G., Pluijm, S. M. F., Jonker, C., Deeg, D. J. H., Lomecky, M. Z., & Lips, P. (2003). Insulin-like growth factor I (IGF-I) and cognitive decline in older persons. *Neurobiology of Aging*, *24*(4), 573–581.
Dincheva, I., Glatt, C. E., & Lee, F. S. (2012). Impact of the BDNF Val66Met Polymorphism on Cognition: Implications for Behavioral Genetics. *The Neuroscientist: A Review Journal Bringing Neurobiology, Neurology and Psychiatry*. http://doi.org/10.1177/1073858411431646
Ding, Q., Vaynman, S., Akhavan, M., Ying, Z., & Gomez-Pinilla, F. (2006). Insulin-like growth factor I interfaces with brain-derived neurotrophic factor-mediated synaptic plasticity to modulate aspects of exercise-induced cognitive function. *Neuroscience*, *140*(3), 823–33. http://doi.org/10.1016/j.neuroscience.2006.02.084
Diorio, C., Brisson, J., Bérubé, S., & Pollak, M. (2008). Genetic polymorphisms involved in insulin-like growth factor (IGF) pathway in relation to mammographic breast density and IGF levels. *Cancer Epidemiol Biomarkers Prev*, *17*(4), 880–8. http://doi.org/10.1158/1055-9965.EPI-07-2500
Egan, M. F., Goldberg, T. E., Kolachana, B. S., Callicott, J. H., Mazzanti, C. M., Straub, R. E., … Weinberger, D. R. (2001). Effect of COMT Val108/158 Met genotype on frontal lobe function and risk for schizophrenia. *Proc Natl Acad Sci USA*, *98*(12), 6917–22. http://doi.org/10.1073/pnas.111134598
Egan, M. F., Kojima, M., Callicott, J. H., Goldberg, T. E., Kolachana, B. S., Bertolino, A., … Weinberger, D. R. (2003). The BDNF val66met polymorphism affects activity-dependent secretion of BDNF and human memory and hippocampal function. *Cell*, *112*(2), 257–69.
Erickson, K. I., Banducci, S. E., Weinstein, A. M., MacDonald, A. W., Ferrell, R. E., Halder, I., … Manuck, S. B. (2013). The Brain-Derived Neurotrophic Factor Val66Met Polymorphism Moderates an Effect of Physical Activity on Working Memory Performance. *Psychological Science*, *24*(9), 1770–1779. http://doi.org/10.1177/0956797613480367
Erickson, K. I., & Kramer, A. F. (2009). Aerobic exercise effects on cognitive and neural plasticity in older adults. *British Journal of Sports Medicine*, *43*(1), 22–4. http://doi.org/10.1136/bjsm.2008.052498

Erickson, K. I., Prakash, R. S., Kim, J. S., Sutton, B. P., Colcombe, S. J., & Kramer, A. F. (2009). Top-down attentional control in spatially coincident stimuli enhances activity in both task-relevant and task-irrelevant regions of cortex. *Behavioural Brain Research*, *197*(1), 186–197. http://doi.org/10.1016/j.bbr.2008.08.028

Erickson, K. I., Prakash, R. S., Voss, M. W., Chaddock, L., Heo, S., McLaren, M., … Kramer, A. F. (2010). Brain-derived neurotrophic factor is associated with age-related decline in hippocampal volume. *J Neurosci*, *30*(15), 5368–75. http://doi.org/10.1523/JNEUROSCI.6251-09.2010

Erickson, K. I., Prakash, R. S., Voss, M. W., Chaddock, L., Hu, L., Morris, K. S., … Kramer, A. F. (2009). Aerobic fitness is associated with hippocampal volume in elderly humans. *Hippocampus*, *19*(10), 1030–9. http://doi.org/10.1002/hipo.20547

Erixon-Lindroth, N., Farde, L., Wahlin, T.-B. R., Sovago, J., Halldin, C., & Bäckman, L. (2005). The role of the striatal dopamine transporter in cognitive aging. *Psychiatry Res*, *138*(1), 1–12. http://doi.org/10.1016/j.pscychresns.2004.09.005

Etnier, J. L., Caselli, R. J., Reiman, E. M., Alexander, G. E., Sibley, B. A., Tessier, D., & Mclemore, E. C. (2007). Cognitive performance in older women relative to ApoE-epsilon4 genotype and aerobic fitness. *Medicine & Science in Sports & Exercise*, *39*(1), 199–207. http://doi.org/10.1249/01.mss.0000239399.85955.5e

Etnier, J. L., Labban, J. D., Karper, W. B., Wideman, L., Piepmeier, A. T., Shih, C. H., … Rulison, K. L. (2015). Innovative Research Exploring the Effects of Physical Activity and Genetics on Cognitive Performance in Community-Based Older Adults. *Journal of Aging and Physical Activity*, *23*(4), 559–568. http://doi.org/10.1123/japa.2014-0221

Etnier, J. ., Salazar, W., Landers, D. ., Petruzzello, S. ., Han, M., & Nowell, P. (1997). The influence of physical fitness and exercise upon cognitive functioning: A meta-analysis. *Journal of Sport & Exercise Psychology*.

Fan, J., Wu, Y., Fossella, J. A., & Posner, M. I. (2001). Assessing the heritability of attentional networks. *BMC Neurosci*, *2*, 14.

Fernandez, A. M., & Torres-Alemán, I. (2012). The many faces of insulin-like peptide signalling in the brain. *Nature Reviews Neuroscience*, *13*(4), 225–239. http://doi.org/10.1038/nrn3209

Floresco, S. B., & Phillips, A. G. (2001). Delay-dependent modulation of memory retrieval by infusion of a dopamine D1 agonist into the rat medial prefrontal cortex. *Behavioral Neuroscience*, *115*(4), 934–9.

Froesch, E. R., Schmid, C., Schwander, J., & Zapf, J. (1985). Actions of insulin-like growth factors. *Annual Review of Physiology*, *47*, 443–467. http://doi.org/10.1146/annurev.ph.47.030185.002303

Garcia-Estrada, J., Garcia-Segura, L. M., & Torres-Aleman, I. (1992). Expression of insulin-like growth factor I by astrocytes in response to injury. *Brain Research*, *592*(1-2), 343–347.

Gilliam, P. E., Spirduso, W. W., Martin, T. P., Walters, T. J., Wilcox, R. E., & Farrar, R. P. (1984). The effects of exercise training on [3H]-spiperone binding in rat striatum. *Pharmacology, Biochemistry, and Behavior*, *20*(6), 863–867.

Gogos, J. A., Morgan, M., Luine, V., Santha, M., Ogawa, S., Pfaff, D., & Karayiorgou, M. (1998). Catechol-O-methyltransferase-deficient mice exhibit sexually dimorphic changes in catecholamine levels and behavior. *Proceedings of the National Academy of Sciences of the United States of America*, *95*(17), 9991–9996.

Goldberg, T. E., Egan, M. F., Gscheidle, T., Coppola, R., Weickert, T., Kolachana, B. S., … Weinberger, D. R. (2003). Executive subprocesses in working memory: relationship to catechol-O-methyltransferase Val158Met genotype and schizophrenia. *Arch Gen Psychiatry*, *60*(9), 889–96. http://doi.org/10.1001/archpsyc.60.9.889

Goldberg, T. E., & Weinberger, D. R. (2004). Genes and the parsing of cognitive processes. *Trends Cogn Sci (Regul Ed)*, *8*(7), 325–35. http://doi.org/10.1016/j.tics.2004.05.011

Gomez-Pinilla, F., & Hillman, C. (2013). The Influence of Exercise on Cognitive Abilities. In R. Terjung (Ed.), *Comprehensive Physiology*. Hoboken, NJ, USA: John Wiley & Sons, Inc. Retrieved from http://doi.wiley.com/10.1002/cphy.c110063

Greene, C. M., Bellgrove, M. A., Gill, M., & Robertson, I. H. (2009). Noradrenergic genotype predicts lapses in sustained attention. *Neuropsychologia*, *47*(2), 591–4. http://doi.org/10.1016/j.neuropsychologia.2008.10.003

Greene, C. M., Braet, W., Johnson, K. A., & Bellgrove, M. A. (2008). Imaging the genetics of executive function. *Biological Psychology*, *79*(1), 30–42. http://doi.org/10.1016/j.biopsycho.2007.11.009

Greenwood, P. M., Lin, M.-K., Sundararajan, R., Fryxell, K. J., & Parasuraman, R. (2014). Healthy aging increases the cognitive effects of two genes that influence extracellular dopamine. *Psychology and Aging*, *29*(2), 363–373. http://doi.org/10.1037/a0036109

Greenwood, P. M., & Parasuraman, R. (2003). Normal genetic variation, cognition, and aging. *Behav Cogn Neurosci Rev*, *2*(4), 278–306. http://doi.org/10.1177/1534582303260641

Greenwood, P. M., Sundararajan, R., Lin, M.-K., Kumar, R., Fryxell, K. J., & Parasuraman, R. (2009). Both a nicotinic single nucleotide polymorphism (SNP) and a noradrenergic SNP modulate working memory performance when attention is manipulated. *J Cogn Neurosci*, *21*(11), 2139–53. http://doi.org/10.1162/jocn.2008.21164

Guan, J., Williams, C. E., Skinner, S. J., Mallard, E. C., & Gluckman, P. D. (1996). The effects of insulin-like growth factor (IGF)-1, IGF-2, and des-IGF-1 on neuronal loss after hypoxic-ischemic brain injury in adult rats: evidence for a role for IGF binding proteins. *Endocrinology*, *137*(3), 893–898.

Guan, J., Williams, C., Gunning, M., Mallard, C., & Gluckman, P. (1993). The effects of IGF-1 treatment after hypoxic-ischemic brain injury in adult rats. *Journal of Cerebral Blood Flow and Metabolism: Official Journal of the International Society of Cerebral Blood Flow and Metabolism*, *13*(4), 609–616. http://doi.org/10.1038/jcbfm.1993.79

Gunstad, J., Benitez, A., Smith, J., Glickman, E., Spitznagel, M. B., Alexander, T., … Murray, L. (2008). Serum Brain-Derived Neurotrophic Factor Is Associated With Cognitive Function in Healthy Older Adults. *Journal of Geriatric Psychiatry and Neurology*, *21*(3), 166–170. http://doi.org/10.1177/0891988708316860

Hall, K., Hilding, A., & Thorén, M. (1999). Determinants of circulating insulin-like growth factor-I. *Journal of Endocrinological Investigation*, *22*(5 Suppl), 48–57.

Han, V. K. (1995). Is the central nervous system a target for growth hormone and insulin-like growth factors? *Acta Paediatrica (Oslo, Norway: 1992). Supplement*, *411*, 3–8.

Hare, M. P., & Palumbi, S. R. (1999). The accuracy of heterozygous base calling from diploid sequence and resolution of haplotypes using allele-specific sequencing. *Molecular Ecology*, *8*(10), 1750–1752.

Hariri, A. R., Goldberg, T. E., Mattay, V. S., Kolachana, B. S., Callicott, J. H., Egan, M. F., & Weinberger, D. R. (2003). Brain-derived neurotrophic factor val66met polymorphism affects human memory-related hippocampal activity and predicts memory performance. *J Neurosci*, *23*(17), 6690–4.

Harrela, M., Koistinen, H., Kaprio, J., Lehtovirta, M., Tuomilehto, J., Eriksson, J., … Seppälä, M. (1996). Genetic and environmental components of interindividual variation in circulating levels of IGF-I, IGF-II, IGFBP-1, and IGFBP-3. *The Journal of Clinical Investigation*, *98*(11), 2612–2615. http://doi.org/10.1172/JCI119081

Harris, S. E., & Deary, I. J. (2011). The genetics of cognitive ability and cognitive ageing in healthy older people. *Trends in Cognitive Sciences*. http://doi.org/10.1016/j.tics.2011.07.004

Hedden, T., & Gabrieli, J. D. E. (2004). Insights into the ageing mind: a view from cognitive neuroscience. *Nat Rev Neurosci*, *5*(2), 87–96. http://doi.org/10.1038/nrn1323

Hess, C., Reif, A., Strobel, A., Boreatti-Hümmer, A., Heine, M., Lesch, K.-P., & Jacob, C. P. (2009). A functional dopamine-beta-hydroxylase gene promoter polymorphism is associated with impulsive personality styles, but not with affective disorders. *J Neural Transm*, *116*(2), 121–30. http://doi.org/10.1007/s00702-008-0138-0

Hillman, C. H., Weiss, E. P., Hagberg, J. M., & Hatfield, B. D. (2002). The relationship of age and cardiovascular fitness to cognitive and motor processes. *Psychophysiology*, *39*(3), 303–12. http://doi.org/10.1017.S0048577201393058

Hof, P. R., & Morrison, J. H. (2004). The aging brain: morphomolecular senescence of cortical circuits. *Trends Neurosci*, *27*(10), 607–13. http://doi.org/10.1016/j.tins.2004.07.013

Hong, Y., Pedersen, N. L., Brismar, K., Hall, K., & de Faire, U. (1996). Quantitative genetic analyses of insulin-like growth factor I (IGF-I), IGF-binding protein-1, and insulin levels in middle-aged and elderly twins. *The Journal of Clinical Endocrinology and Metabolism*, *81*(5), 1791–1797.

Huuskonen, A., Lappalainen, J., Oksala, N., Santtila, M., HäKkinen, K., KyröLäInen, H., & Atalay, M. (2011). Common Genetic Variation in the IGF1 Associates with Maximal Force Output. *Medicine & Science in Sports & Exercise*, *43*(12), 2368–2374. http://doi.org/10.1249/MSS.0b013e3182220179

Hwang, I. K., Yoo, K.-Y., Jung, B.-K., Cho, J. H., Kim, D.-H., Kang, T.-C., … Won, M. H. (2006). Correlations between neuronal loss, decrease of memory, and decrease expression of brain-derived neurotrophic factor in

the gerbil hippocampus during normal aging. *Experimental Neurology*, *201*(1), 75–83. http://doi.org/10.1016/j.expneurol.2006.02.129

Jack, C. R., Jr, Petersen, R. C., Xu, Y., O'Brien, P. C., Smith, G. E., Ivnik, R. J., … Kokmen, E. (1998). Rate of medial temporal lobe atrophy in typical aging and Alzheimer's disease. *Neurology*, *51*(4), 993–999.

Johansson, M., Mckay, J. D., Wiklund, F., Rinaldi, S., Verheus, M., Gils, C. H. van, … Kaaks, R. (2007). Implications for prostate cancer of insulin-like growth factor-I (IGF-I) genetic variation and circulating IGF-I levels. *Journal of Clinical Endocrinology & Metabolism*, *92*(12), 4820–6. http://doi.org/10.1210/jc.2007-0887

Johnson, M. K., Raye, C. L., Mitchell, K. J., Greene, E. J., & Anderson, A. W. (2003). FMRI evidence for an organization of prefrontal cortex by both type of process and type of information. *Cereb Cortex*, *13*(3), 265–73.

Karoum, F., Chrapusta, S. J., & Egan, M. F. (1994). 3-Methoxytyramine is the major metabolite of released dopamine in the rat frontal cortex: reassessment of the effects of antipsychotics on the dynamics of dopamine release and metabolism in the frontal cortex, nucleus accumbens, and striatum by a simple two pool model. *Journal of Neurochemistry*, *63*(3), 972–979.

Kaufman, A. S., & Kaufman, N. L. (1990). *Kaufman Brief Intelligence Test manual*. Circle Pines, MN: American Guidance Service.

Kleim, J. A., Cooper, N. R., & VandenBerg, P. M. (2002). Exercise induces angiogenesis but does not alter movement representations within rat motor cortex. *Brain Research*, *934*(1), 1–6. http://doi.org/10.1016/S0006-8993(02)02239-4

Kobilo, T., Liu, Q.-R., Gandhi, K., Mughal, M., Shaham, Y., & van Praag, H. (2011). Running is the neurogenic and neurotrophic stimulus in environmental enrichment. *Learning & Memory*, *18*(9), 605–609. http://doi.org/10.1101/lm.2283011

Kopecková, M., Paclt, I., & Goetz, P. (2006). Polymorphisms of dopamine-beta-hydroxylase in ADHD children. *Folia Biol (Praha)*, *52*(6), 194–201.

Kramer, A. F., & Erickson, K. I. (2007). Capitalizing on cortical plasticity: influence of physical activity on cognition and brain function. *Trends Cogn Sci (Regul Ed)*, *11*(8), 342–8. http://doi.org/10.1016/j.tics.2007.06.009

Kramer, A. F., Hahn, S., & Gopher, D. (1999). Task coordination and aging: explorations of executive control processes in the task switching paradigm. *Acta Psychol (Amst)*, *101*(2-3), 339–78.

Langley, L. K., & Madden, D. J. (2000). Functional neuroimaging of memory: implications for cognitive aging. *Microsc Res Tech*, *51*(1), 75–84. http://doi.org/10.1002/1097-0029(20001001)51:1<75::AID-JEMT8>3.0.CO;2-6

Lee, T., Henry, J. D., Trollor, J. N., & Sachdev, P. S. (2010). Genetic influences on cognitive functions in the elderly: A selective review of twin studies. *Brain Research Reviews*, *64*(1), 1–13. http://doi.org/10.1016/j.brainresrev.2010.02.001

Licht, C. M. M., van Turenhout, L. C., Deijen, J. B., Koppes, L. L. J., van Mechelen, W., Twisk, J. W. R., & Drent, M. L. (2014). The Association between IGF-1 Polymorphisms, IGF-1 Serum Levels, and Cognitive Functions in Healthy Adults: The Amsterdam Growth and Health Longitudinal Study. *International Journal of Endocrinology*, *2014*, 1–8. http://doi.org/10.1155/2014/181327

Li, L., Oppenheim, R. W., Lei, M., & Houenou, L. J. (1994). Neurotrophic agents prevent motoneuron death following sciatic nerve section in the neonatal mouse. *Journal of Neurobiology*, *25*(7), 759–766. http://doi.org/10.1002/neu.480250702

Lipsky, R. H., Xu, K., Zhu, D., Kelly, C., Terhakopian, A., Novelli, A., & Marini, A. M. (2001). Nuclear factor kappaB is a critical determinant in N-methyl-D-aspartate receptor-mediated neuroprotection. *Journal of Neurochemistry*, *78*(2), 254–264.

Li, S.-C., & Rieckmann, A. (2014). Neuromodulation and aging: implications of aging neuronal gain control on cognition. *Current Opinion in Neurobiology*, *29*, 148–158. http://doi.org/10.1016/j.conb.2014.07.009

Lista, I., & Sorrentino, G. (2010). Biological mechanisms of physical activity in preventing cognitive decline. *Cell Mol Neurobiol*, *30*(4), 493–503. http://doi.org/10.1007/s10571-009-9488-x

Liu, L., Forsell, C., Lilius, L., Axelman, K., Corder, E. H., & Lannfelt, L. (1996). Allelic association but only weak evidence for linkage to the apolipoprotein E locus in late-onset Swedish Alzheimer families. *Am J Med Genet*, *67*(3), 306–11. http://doi.org/10.1002/(SICI)1096-8628(19960531)67:3<306::AID-AJMG7>3.0.CO;2-K

Lommatzsch, M., Zingler, D., Schuhbaeck, K., Schloetcke, K., Zingler, C., Schuff-Werner, P., & Virchow, J. C. (2005). The impact of age, weight and gender on BDNF levels in human platelets and plasma. *NBA*, *26*(1), 115–23. http://doi.org/10.1016/j.neurobiolaging.2004.03.002

Lotta, T., Vidgren, J., Tilgmann, C., Ulmanen, I., Melén, K., Julkunen, I., & Taskinen, J. (1995). Kinetics of human soluble and membrane-bound catechol O-methyltransferase: a revised mechanism and description of the thermolabile variant of the enzyme. *Biochemistry*, *34*(13), 4202–10.

MacRae, P. G., Spirduso, W. W., Walters, T. J., Farrar, R. P., & Wilcox, R. E. (1987). Endurance training effects on striatal D2 dopamine receptor binding and striatal dopamine metabolites in presenescent older rats. *Psychopharmacology*, *92*(2), 236–240.

Malhotra, A. K., Kestler, L. J., Mazzanti, C., Bates, J. A., Goldberg, T., & Goldman, D. (2002). A functional polymorphism in the COMT gene and performance on a test of prefrontal cognition. *Am J Psychiatry*, *159*(4), 652–4.

Markowska, A. L., Mooney, M., & Sonntag, W. E. (1998). Insulin-like growth factor-1 ameliorates age-related behavioral deficits. *Neuroscience*, *87*(3), 559–69.

Mather, K. A., Kwok, J. B., Armstrong, N., & Sachdev, P. S. (2014). The role of epigenetics in cognitive ageing: Epigenetics and cognitive ageing. *International Journal of Geriatric Psychiatry*, *29*(11), 1162–1171. http://doi.org/10.1002/gps.4183

Mattay, V. S., & Goldberg, T. E. (2004). Imaging genetic influences in human brain function. *Curr Opin Neurobiol*, *14*(2), 239–47. http://doi.org/10.1016/j.conb.2004.03.014

Mattay, V. S., Goldberg, T. E., Fera, F., Hariri, A. R., Tessitore, A., Egan, M. F., … Weinberger, D. R. (2003). Catechol O-methyltransferase val158-met genotype and individual variation in the brain response to amphetamine. *Proc Natl Acad Sci USA*, *100*(10), 6186–91. http://doi.org/10.1073/pnas.0931309100

Mcauley, E., Kramer, A. F., & Colcombe, S. J. (2004). Cardiovascular fitness and neurocognitive function in older adults: a brief review. *Brain Behav Immun*, *18*(3), 214–20.

Meeusen, R. (2005). Exercise and the brain: insight in new therapeutic modalities. *Ann Transplant*, *10*(4), 49–51.

Meeusen, R., Smolders, I., Sarre, S., de Meirleir, K., Keizer, H., Serneels, M., … Michotte, Y. (1997). Endurance training effects on neurotransmitter release in rat striatum: an in vivo microdialysis study. *Acta Physiologica Scandinavica*, *159*(4), 335–341.

Milham, M. P., Banich, M. T., Webb, A., Barad, V., Cohen, N. J., Wszalek, T., & Kramer, A. F. (2001). The relative involvement of anterior cingulate and prefrontal cortex in attentional control depends on nature of conflict. *Brain Research. Cognitive Brain Research*, *12*(3), 467–473.

Miyajima, F., Ollier, W., Mayes, A., Jackson, A., Thacker, N., Rabbitt, P., … Payton, A. (2008). Brain-derived neurotrophic factor polymorphism Val66Met influences cognitive abilities in the elderly. *Genes Brain Behav*, *7*(4), 411–7. http://doi.org/10.1111/j.1601-183X.2007.00363.x

Miyake, A., Friedman, N. P., Emerson, M. J., Witzki, A. H., Howerter, A., & Wager, T. D. (2000). The unity and diversity of executive functions and their contributions to complex "Frontal Lobe" tasks: a latent variable analysis. *Cogn Psychol*, *41*(1), 49–100. http://doi.org/10.1006/cogp.1999.0734

Monchi, O., Hyun Ko, J., & Strafella, A. P. (2006). Striatal dopamine release during performance of executive functions: A [11C] raclopride PET study. *NeuroImage*, *33*(3), 907–912. http://doi.org/10.1016/j.neuroimage.2006.06.058

Morley, J. E., Kaiser, F., Raum, W. J., Perry, H. M., 3rd, Flood, J. F., Jensen, J., … Roberts, E. (1997). Potentially predictive and manipulable blood serum correlates of aging in the healthy human male: progressive decreases in bioavailable testosterone, dehydroepiandrosterone sulfate, and the ratio of insulin-like growth factor 1 to growth hormone. *Proceedings of the National Academy of Sciences of the United States of America*, *94*(14), 7537–7542.

Morrison, J. H., & Hof, P. R. (2007). Life and death of neurons in the aging cerebral cortex. *Int Rev Neurobiol*, *81*, 41–57. http://doi.org/10.1016/S0074-7742(06)81004-4

Nackley, A. G., Shabalina, S. A., Tchivileva, I. E., Satterfield, K., Korchynskyi, O., Makarov, S. S., … Diatchenko, L. (2006). Human catechol-O-methyltransferase haplotypes modulate protein expression by altering mRNA secondary structure. *Science*, *314*(5807), 1930–3. http://doi.org/10.1126/science.1131262

Nagel, I. E., Chicherio, C., Li, S.-C., Oertzen, T. von, Sander, T., Villringer, A., … Lindenberger, U. (2008). Human aging magnifies genetic effects on executive functioning and working memory. *Front. Hum. Neurosci.*, *2*, 1. http://doi.org/10.3389/neuro.09.001.2008

Neeper, S. A., Gómez-Pinilla, F., Choi, J., & Cotman, C. (1995). Exercise and brain neurotrophins. *Nature*, *373*(6510), 109. http://doi.org/10.1038/373109a0
Neeper, S. A., Gómez-Pinilla, F., Choi, J., & Cotman, C. W. (1996). Physical activity increases mRNA for brain-derived neurotrophic factor and nerve growth factor in rat brain. *Brain Research*, *726*(1-2), 49–56. http://doi.org/10.1016/0006-8993(96)00273-9
Nieoullon, A. (2002). Dopamine and the regulation of cognition and attention. *Prog Neurobiol*, *67*(1), 53–83.
Nishijima, T., Piriz, J., Duflot, S., Fernandez, A. M., Gaitan, G., Gomez-Pinedo, U., … Nuñez, A. (2010). Neuronal Activity Drives Localized Blood-Brain-Barrier Transport of Serum Insulin-like Growth Factor-I into the CNS. *Neuron*, *67*(5), 834–846. http://doi.org/10.1016/j.neuron.2010.08.007
O'Sullivan, M., Jones, D. K., Summers, P. E., Morris, R. G., Williams, S. C., & Markus, H. S. (2001). Evidence for cortical "disconnection" as a mechanism of age-related cognitive decline. *Neurology*, *57*(4), 632–8.
Pang, P. T., & Lu, B. (2004). Regulation of late-phase LTP and long-term memory in normal and aging hippocampus: role of secreted proteins tPA and BDNF. *Ageing Research Reviews*, *3*(4), 407–430. http://doi.org/10.1016/j.arr.2004.07.002
Parasuraman, R., Greenwood, P. M., Kumar, R., & Fossella, J. (2005). Beyond heritability: neurotransmitter genes differentially modulate visuospatial attention and working memory. *Psychol Sci*, *16*(3), 200–7. http://doi.org/10.1111/j.0956-7976.2005.00804.x
Park, D. C., & Gutchess, A. H. (2002). Aging, cognition, and culture: a neuroscientific perspective. *Neurosci Biobehav Rev*, *26*(7), 859–67.
Park, D. ., & Gutchess, A. . (2000). Cognitive aging and everyday life. *Cognitive Aging: A Primer*, 217–232.
Pezawas, L., Verchinski, B. A., Mattay, V. S., Callicott, J. H., Kolachana, B. S., Straub, R. E., … Weinberger, D. R. (2004). The brain-derived neurotrophic factor val66met polymorphism and variation in human cortical morphology. *The Journal of Neuroscience: The Official Journal of the Society for Neuroscience*, *24*(45), 10099–10102. http://doi.org/10.1523/JNEUROSCI.2680-04.2004
Plomin, R., & Craig, I. (1997). Human behavioural genetics of cognitive abilities and disabilities. *Bioessays*, *19*(12), 1117–24. http://doi.org/10.1002/bies.950191211
Prakash, R. S., Voss, M. W., Erickson, K. I., & Kramer, A. F. (2015). Physical Activity and Cognitive Vitality. *Annual Review of Psychology*, *66*(1), 769–797. http://doi.org/10.1146/annurev-psych-010814-015249
Ranganath, A., & Jacob, S. N. (2015). Doping the Mind: Dopaminergic Modulation of Prefrontal Cortical Cognition. *The Neuroscientist*. http://doi.org/10.1177/1073858415602850
Raz, N. (2000). Aging of the brain and its impact on cognitive performance: Integration of structural and functional findings. *The Handbook of Aging and Cognition*, *2*, 1–90.
Raz, N., Gunning, F. M., Head, D., Dupuis, J. H., McQuain, J., Briggs, S. D., … Acker, J. D. (1997). Selective aging of the human cerebral cortex observed in vivo: differential vulnerability of the prefrontal gray matter. *Cereb Cortex*, *7*(3), 268–82.
Raz, N., Lindenberger, U., Rodrigue, K. M., Kennedy, K. M., Head, D., Williamson, A., … Acker, J. D. (2005). Regional brain changes in aging healthy adults: general trends, individual differences and modifiers. *Cereb Cortex*, *15*(11), 1676–89. http://doi.org/10.1093/cercor/bhi044
Raz, N., & Lustig, C. (2014). Genetic variants and cognitive aging: Destiny or a nudge? *Psychology and Aging*, *29*(2), 359–362. http://doi.org/10.1037/a0036893
Reinhardt, R. R., & Bondy, C. A. (1994). Insulin-like growth factors cross the blood-brain barrier. *Endocrinology*, *135*(5), 1753–1761.
Renaud, M., Bherer, L., & Maquestiaux, F. (2010). A High Level of Physical Fitness Is Associated With More Efficient Response Preparation in Older Adults. *The Journals of Gerontology Series B: Psychological Sciences and Social Sciences*, *65B*(3), 317–322. http://doi.org/10.1093/geronb/gbq004
Reuter-Lorenz, P. A., Jonides, J., Smith, E. E., Hartley, A., Miller, A., Marshuetz, C., & Koeppe, R. A. (2000). Age differences in the frontal lateralization of verbal and spatial working memory revealed by PET. *J Cogn Neurosci*, *12*(1), 174–87.
Rex, C. S. (2006). Restoration of Long-Term Potentiation in Middle-Aged Hippocampus After Induction of Brain-Derived Neurotrophic Factor. *Journal of Neurophysiology*, *96*(2), 677–685. http://doi.org/10.1152/jn.00336.2006
Rex, C. S., Lin, C.-Y., Kramar, E. A., Chen, L. Y., Gall, C. M., & Lynch, G. (2007). Brain-Derived Neurotrophic Factor Promotes Long-Term Potentiation-Related Cytoskeletal Changes in Adult Hippocampus. *Journal of Neuroscience*, *27*(11), 3017–3029. http://doi.org/10.1523/JNEUROSCI.4037-06.2007

Rollero, A., Murialdo, G., Fonzi, S., Garrone, S., Gianelli, M. V., Gazzerro, E., … Polleri, A. (1998). Relationship between cognitive function, growth hormone and insulin-like growth factor I plasma levels in aged subjects. *Neuropsychobiology*, *38*(2), 73–79.

Roussos, P., Giakoumaki, S. G., & Bitsios, P. (2009). Tolcapone effects on gating, working memory, and mood interact with the synonymous catechol-O-methyltransferase rs4818c/g polymorphism. *Biological Psychiatry*, *66*(11), 997–1004. http://doi.org/10.1016/j.biopsych.2009.07.008

Roussos, P., Giakoumaki, S. G., Pavlakis, S., & Bitsios, P. (2008). Planning, decision-making and the COMT rs4818 polymorphism in healthy males. *Neuropsychologia*, *46*(2), 757–63. http://doi.org/10.1016/j.neuropsychologia.2007.10.009

Rybakowski, J. K., Borkowska, A., Czerski, P. M., Skibińska, M., & Hauser, J. (2003). Polymorphism of the brain-derived neurotrophic factor gene and performance on a cognitive prefrontal test in bipolar patients. *Bipolar Disorders*, *5*(6), 468–472.

Salthouse, T. A., & Miles, J. D. (2002). Aging and time-sharing aspects of executive control. *Mem Cognit*, *30*(4), 572–82.

Savitz, J., Solms, M., & Ramesar, R. (2006). The molecular genetics of cognition: dopamine, COMT and BDNF. *Genes Brain Behav*, *5*(4), 311–28. http://doi.org/10.1111/j.1601-183X.2005.00163.x

Schuit, A. J., Feskens, E. J., Launer, L. J., & Kromhout, D. (2001). Physical activity and cognitive decline, the role of the apolipoprotein e4 allele. *Medicine & Science in Sports & Exercise*, *33*(5), 772–7.

Seamans, J. K., & Yang, C. R. (2004). The principal features and mechanisms of dopamine modulation in the prefrontal cortex. *Prog Neurobiol*, *74*(1), 1–58. http://doi.org/10.1016/j.pneurobio.2004.05.006

Sikström, S. (2007). Computational perspectives on neuromodulation of aging. *Acta Neurochir Suppl*, *97*(Pt 2), 513–8.

Silhol, M., Arancibia, S., Maurice, T., & Tapia-Arancibia, L. (2007). Spatial memory training modifies the expression of brain-derived neurotrophic factor tyrosine kinase receptors in young and aged rats. *Neuroscience*, *146*(3), 962–973. http://doi.org/10.1016/j.neuroscience.2007.02.013

Silhol, M., Arancibia, S., Perrin, D., Maurice, T., Alliot, J., & Tapia-Arancibia, L. (2008). Effect of Aging on Brain-Derived Neurotrophic Factor, proBDNF, and Their Receptors in the Hippocampus of Lou/C Rats. *Rejuvenation Research*, *11*(6), 1031–1040. http://doi.org/10.1089/rej.2008.0791

Simmons, D. A., Rex, C. S., Palmer, L., Pandyarajan, V., Fedulov, V., Gall, C. M., & Lynch, G. (2009). Up-regulating BDNF with an ampakine rescues synaptic plasticity and memory in Huntington's disease knockin mice. *Proceedings of the National Academy of Sciences of the United States of America*, *106*(12), 4906–4911. http://doi.org/10.1073/pnas.0811228106

Slifstein, M., Kolachana, B., Simpson, E. H., Tabares, P., Cheng, B., Duvall, M., … Abi-Dargham, A. (2008). COMT genotype predicts cortical-limbic D1 receptor availability measured with [11C]NNC112 and PET. *Mol Psychiatry*, *13*(8), 821–7. http://doi.org/10.1038/mp.2008.19

Smith, P. J., Blumenthal, J. A., Hoffman, B. M., Cooper, H., Strauman, T. A., Welsh-Bohmer, K., … Sherwood, A. (2010). Aerobic exercise and neurocognitive performance: a meta-analytic review of randomized controlled trials. *Psychosomatic Medicine*, *72*(3), 239–52. http://doi.org/10.1097/PSY.0b013e3181d14633

Söderlund, D., Canto, P., & Méndez, J. P. (2002). Identification of three novel mutations in the KAL1 gene in patients with Kallmann syndrome. *The Journal of Clinical Endocrinology and Metabolism*, *87*(6), 2589–2592.

Spiegel, A. M., Sewal, A. S., & Rapp, P. R. (2014). Epigenetic contributions to cognitive aging: disentangling mindspan and lifespan. *Learning & Memory*, *21*(10), 569–574. http://doi.org/10.1101/lm.033506.113

Spirduso, W. W. (1975). Reaction and Movement Time as a Function of age and Physical Activity Level. *Journal of Gerontology*, *30*(4), 435–440. http://doi.org/10.1093/geronj/30.4.435

Strong, R. (1998). Neurochemical changes in the aging human brain: implications for behavioral impairment and neurodegenerative disease. *Geriatrics*, *53 Suppl 1*, S9–12.

Stroop, J. R. (1935). Studies of interference in serial verbal reactions. *Journal of Experimental Psychology*, *18*(6), 643–662. http://doi.org/10.1037/h0054651

Stroth, S., Reinhardt, R. K., Thöne, J., Hille, K., Schneider, M., Härtel, S., … Spitzer, M. (2010). Impact of aerobic exercise training on cognitive functions and affect associated to the COMT polymorphism in young adults. *Neurobiology of Learning and Memory*, *94*(3), 364–72. http://doi.org/10.1016/j.nlm.2010.08.003

Swan, G. E., & Carmelli, D. (2002). Evidence for Genetic Mediation of Executive Control: A Study of Aging Male Twins. *The Journals of Gerontology Series B: Psychological Sciences and Social Sciences*, *57*(2), P133–P143. http://doi.org/10.1093/geronb/57.2.P133

Trejo, J. L., Carro, E., & Torres-Aleman, I. (2001). Circulating insulin-like growth factor I mediates exercise-induced increases in the number of new neurons in the adult hippocampus. *J Neurosci*, *21*(5), 1628–34.

Trejo, J. L., Llorens-Martín, M. V., & Torres-Alemán, I. (2008). The effects of exercise on spatial learning and anxiety-like behavior are mediated by an IGF-I-dependent mechanism related to hippocampal neurogenesis. *Mol Cell Neurosci*, *37*(2), 402–11. http://doi.org/10.1016/j.mcn.2007.10.016

Tunbridge, E. M., Harrison, P. J., & Weinberger, D. R. (2006). Catechol-o-methyltransferase, cognition, and psychosis: Val158Met and beyond. *Biological Psychiatry*, *60*(2), 141–51. http://doi.org/10.1016/j.biopsych.2005.10.024

van Dam, P. S., & Aleman, A. (2004). Insulin-like growth factor-I, cognition and brain aging. *European Journal of Pharmacology*, *490*(1-3), 87–95. http://doi.org/10.1016/j.ejphar.2004.02.047

van Dam, P. S., Aleman, A., de Vries, W. R., Deijen, J. B., van der Veen, E. A., de Haan, E. H., & Koppeschaar, H. P. (2000). Growth hormone, insulin-like growth factor I and cognitive function in adults. *Growth Hormone & IGF Research: Official Journal of the Growth Hormone Research Society and the International IGF Research Society*, *10 Suppl B*, S69–73.

Van der Linden, M., Brédart, S., & Beerten, A. (1994). Age-related differences in updating working memory. *Br J Psychol*, *85 (Pt 1)*, 145–52.

van Praag, H., Christie, B. R., Sejnowski, T. J., & Gage, F. H. (1999). Running enhances neurogenesis, learning, and long-term potentiation in mice. *Proceedings of the National Academy of Sciences of the United States of America*, *96*(23), 13427–13431.

van Praag, H., Kempermann, G., & Gage, F. H. (1999). Running increases cell proliferation and neurogenesis in the adult mouse dentate gyrus. *Nature Neuroscience*, *2*(3), 266–270. http://doi.org/10.1038/6368

Vaynman, S., & Gomez-Pinilla, F. (2006). Revenge of the "sit": how lifestyle impacts neuronal and cognitive health through molecular systems that interface energy metabolism with neuronal plasticity. *J. Neurosci. Res.*, *84*(4), 699–715. http://doi.org/10.1002/jnr.20979

Vaynman, S., Ying, Z., & Gomez-Pinilla, F. (2004). Hippocampal BDNF mediates the efficacy of exercise on synaptic plasticity and cognition. *Eur J Neurosci*, *20*(10), 2580–90. http://doi.org/10.1111/j.1460-9568.2004.03720.x

Verhaeghe, J., Loos, R., Vlietinck, R., Herck, E. V., van Bree, R., & Schutter, A. M. (1996). C-peptide, insulin-like growth factors I and II, and insulin-like growth factor binding protein-1 in cord serum of twins: genetic versus environmental regulation. *American Journal of Obstetrics and Gynecology*, *175*(5), 1180–1188.

Verhaeghen, P., & Cerella, J. (2002). Aging, executive control, and attention: a review of meta-analyses. *Neurosci Biobehav Rev*, *26*(7), 849–57.

Verhaeghen, P., Steitz, D. W., Sliwinski, M. J., & Cerella, J. (2003). Aging and dual-task performance: a meta-analysis. *Psychology and Aging*, *18*(3), 443–60. http://doi.org/10.1037/0882-7974.18.3.443

Verheus, M., Mckay, J. D., Kaaks, R., Canzian, F., Biessy, C., Johansson, M., … Gils, C. H. van. (2008). Common genetic variation in the IGF-1 gene, serum IGF-I levels and breast density. *Breast Cancer Res Treat*, *112*(1), 109–22. http://doi.org/10.1007/s10549-007-9827-x

Vitiello, M., Moe, K., Merriam, G., Mazzoni, G., Buchner, D., & Schwartz, R. (2006). Growth hormone releasing hormone improves the cognition of healthy older adults. *Neurobiology of Aging*, *27*(2), 318–323. http://doi.org/10.1016/j.neurobiolaging.2005.01.010

Voelcker-Rehage, C., Jeltsch, A., Godde, B., Becker, S., & Staudinger, U. M. (2015). COMT gene polymorphisms, cognitive performance, and physical fitness in older adults. *Psychology of Sport and Exercise*, *20*, 20–28. http://doi.org/10.1016/j.psychsport.2015.04.001

Volkow, N. D., Gur, R. C., Wang, G. J., Fowler, J. S., Moberg, P. J., Ding, Y. S., … Logan, J. (1998). Association between decline in brain dopamine activity with age and cognitive and motor impairment in healthy individuals. *The American Journal of Psychiatry*, *155*(3), 344–349. http://doi.org/10.1176/ajp.155.3.344

Voss, M. W., Erickson, K. I., Prakash, R. S., Chaddock, L., Malkowski, E., Alves, H., … Kramer, A. F. (2010). Functional connectivity: a source of variance in the association between cardiorespiratory fitness

and cognition? *Neuropsychologia*, *48*(5), 1394–406. http://doi.org/10.1016/j.neuropsychologia.2010.01.005

Wang, X., Kong, L., Zhang, G., Sun, M., & Geller, A. I. (2005). Targeted gene transfer to nigrostriatal neurons in the rat brain by helper virus-free HSV-1 vector particles that contain either a chimeric HSV-1 glycoprotein C-GDNF or a gC-BDNF protein. *Brain Research. Molecular Brain Research*, *139*(1), 88–102. http://doi.org/10.1016/j.molbrainres.2005.05.029

West, R. L. (1996). An application of prefrontal cortex function theory to cognitive aging. *Psychological Bulletin*, *120*(2), 272–92.

Winter, B., Breitenstein, C., Mooren, F. C., Voelker, K., Fobker, M., Lechtermann, A., … Knecht, S. (2007). High impact running improves learning. *Neurobiology of Learning and Memory*, *87*(4), 597–609. http://doi.org/10.1016/j.nlm.2006.11.003

Yamaguchi, F., Itano, T., Miyamoto, O., Janjua, N. A., Ohmoto, T., Hosokawa, K., & Hatase, O. (1991). Increase of extracellular insulin-like growth factor I (IGF-I) concentration following electrolytical lesion in rat hippocampus. *Neuroscience Letters*, *128*(2), 273–276.

Yankner, B. A., Lu, T., & Loerch, P. (2008). The aging brain. *Annu Rev Pathol*, *3*, 41–66. http://doi.org/10.1146/annurev.pathmechdis.2.010506.092044

Zabetian, C. P., Anderson, G. M., Buxbaum, S. G., Elston, R. C., Ichinose, H., Nagatsu, T., … Cubells, J. F. (2001). A quantitative-trait analysis of human plasma-dopamine beta-hydroxylase activity: evidence for a major functional polymorphism at the DBH locus. *American Journal of Human Genetics*, *68*(2), 515–522. http://doi.org/10.1086/318198

Zabetian, C. P., Buxbaum, S. G., Elston, R. C., Köhnke, M. D., Anderson, G. M., Gelernter, J., & Cubells, J. F. (2003). The structure of linkage disequilibrium at the DBH locus strongly influences the magnitude of association between diallelic markers and plasma dopamine beta-hydroxylase activity. *American Journal of Human Genetics*, *72*(6), 1389–1400. http://doi.org/10.1086/375499

Ziegenhorn, A. A., Schulte-Herbrüggen, O., Danker-Hopfe, H., Malbranc, M., Hartung, H.-D., Anders, D., … Hellweg, R. (2007). Serum neurotrophins–a study on the time course and influencing factors in a large old age sample. *Neurobiol Aging*, *28*(9), 1436–45. http://doi.org/10.1016/j.neurobiolaging.2006.06.011